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QUAESTIONES ENTOMOLOGICAE

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Editorial — Words, words, words

The first edition of the World List of Scientific Periodicals, published in 1921, listed 25,000 titles. The second edition in 1934 listed more than 36,000; the third edition in 1952 listed more than 50,000. The fourth edition now appearing lists over 60,000, despite the fact that "some 10,000 titles included in the third edition have been left out as being of social or commercial rather than scientific interest". Most periodicals have recently waxed fat, so that one may estimate 25 years as the time in which the flow of scientific literature doubles itself.

By comparison with science as a whole, the growth of entomological literature seems somewhat pedestrian; the Insecta portion of the Zoological Record listed 1970 titles of papers in 1921 and 4024 in 1953. The applied literature, as represented by the Review of Applied Entomology has been, somewhat surprisingly, growing more slowly than this, so that one may estimate 35 years for the entomological literature to double itself. Even so the 25,229 entomological articles listed in Horn and Schenkling as published from the beginning of history until the end of 1863, at current rates would be produced in about four years, and the total number of scientific papers now published in the field of entomology must exceed a quarter of a million. One may suspect, however, a shrinkage in the mean length of papers under the joint influence of mounting page charges and the philosophy of "publish or perish" coupled with the waning ability of administrators to judge publications by anything beyond their number.

Some may say that in this situation a new periodical should be offered with an apology - if at all. But if we would slow down the march of science, we must stop research before it has begun, not lose the results of it when it is all but finished. Certainly we must see to it

that we do not produce new facts faster than we can assimilate them into generalizations, although this process calls for that very breadth of outlook which the literature flood makes it difficult for us to achieve. If we can no longer achieve individual breadth, we must provide for composite breadth by facilitating diversity of training and the unusual combination of subjects.

If we stagger under the impact of a swelling literature, before we call for a slow down in research we should remind ourselves that a quarter of a million entomological papers only represents less than one per described species of beetle, and that more than half the species of insects remain to be found and described.

If then, this growth of the literature must go on, what can we do to keep abreast of it? A great many things: fight the trend to shorter papers, which has now reached the ridiculous stage when an index card for a paper may be larger than the content of the paper itself. It costs more in time, money, and effort, to produce, file, store, retrieve, and read ten one page papers than one ten page paper. Publish in the most appropriate periodical from the subject viewpoint; publish promptly; index and abstract everything diversely; and make full use of modern techniques such as microforms, punch cards, and even computers. It may seem redundant to say that material should be published once only, yet how often do we find it difficult to avoid duplicate publication of material from the proceedings of a meeting, and how often is this due to inappropriate publication in the first place? A marriage between microcards and punch cards is long overdue; if sufficiently prolific, the hybrid offspring would be of inestimable value to the bibliographer.

There are signs that things are beginning to move in this direction; perhaps this periodical is one of them. But one may question whether the move is fast enough to get us out of chaos: movable type, despite its name, is conservative stuff.

Despite our concern for the future, we should be both remiss and churlish to enter 1965 without a backward glance to 1865 and the beginning of the Zoological Record. Let us pay both dollars and respect to our venerable abstracting and indexing services - in no other field of endeavour is continuity more important. I wonder whether any other branch of science is as fortunate as entomology with its Hagen, Horn and Schenkling, and Zoological Record. Many complain of the increasing delay in publication of successive volumes of Zoological Record, but how many of the complainants have ever attempted a similar task? And whose fault is this? As Günther pointed out in his preface to volume one in August 1865, many journals of learned societies which would carry the date 1864 on their title pages, had still not appeared; but here we are treading on dangerous ground. We regard it as a most fortunate and propitious honour, to commence publication in the year in which the Zoological Record celebrates its centenary.

Brian Hocking

EFFECTS OF REPELLENTS ON MOSQUITO BEHAVIOR

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The behavior of Aedes aegypti L. and other species of Aedes in relation to repellent chemicals was studied. The repellents used were dimethyl phthalate, ethyl hexanediol, N, N-diethyl metatoluamide and indalone. The effect of these repellents on the behaviour of mosquitoes was studied firstly by placing the repellents on selected parts of the environment and secondly by painting them on parts of mosquitoes themselves where chemoreceptors are known to occur, such as the antennae, labium, and tarsi. The aspects of behavior studied were: feeding on blood and on sugars, mating, oviposition, the reactions to wind, geotaxis and orientation to centrifugal force, and the visual response to black stripes. All these aspects of behavior are affected significantly by repellents. Dimethyl phthalate has the greatest effect of the four repellents on blood feeding behavior when they are painted on the tarsal receptors and the smallest effect when they are painted on the receptors of the antennae and the labium.

The experiments provided some understanding of the mode of action of insect repellents. They suggest that repellents interfere with normal behavior perhaps by blocking the olfactory receptors mediating attraction to food and the contact chemoreceptors invoking feeding on blood and those used in the selection of oviposition sites. The experiments show that mechanoreceptors effecting orientation to gravity and air flow and visual receptors effecting orientation to black stripes are also interfered with by repellents. There is also some evidence that repellents block the thermoreceptors which may mediate piercing for feeding on blood and perhaps auditory organs involved in mating. The only receptors which the repellents do not appear to interfere with seem to be those of the common chemical sense.

INTRODUCTION

The discovery of the transmission of malarial parasites by Ross (1898) and the discovery by Walter Reed and his collaborators that yellow fever was transmitted by Aedes aegypti led to the realization of the importance of mosquitoes as carrier of disease. Repellents being a cheap and efficient means of individual protection, many workers studied their effects mainly against the blood feeding behavior of insects. Kalmus & Hocking (1960), however, studied some other aspects of behavior as well. I have studied the behavior of Aedes aegypti in the presence of repellents not only in relation to blood feeding but also in relation to sugar feeding, mating, oviposition, geotaxis, wind direction and speeds, and visual responses to black stripes. The repellents used were: dimethyl phthalate, indalone, diethyl toluamide, and Rutger's 612. The first two are esters, the third an amide and the last named an alcohol. These are compounds of low volatility and moderate molecular weight. They are insoluble or only very slightly soluble in water but are miscible with alcohol and ether. Their physical and chemical properties are listed in table 1.

TABLE 1 - Chemical and physical properties of the repellents used in the study of behavior of *Aedes*.

Common Name	Chemical Name	Mol. Wt.	Boiling Point	Solubility and Miscibility
Dimethyl phthalate	dimethyl benzene-ortho-dicarboxylate	194.18	285°C	0.43% w/w soluble in water
Indalone	n-butyl mesityl-oxide oxalate	226.26	113°C	Insoluble in water; miscible with alcohol
Diethyl toluamide	N, N-diethyl m-toluamide	191	111°C at 1mm	Insoluble in water; miscible with alcohol
Rutger's 612	2-ethyl, 1, 3-hexanediol	146.22	244°C	Slightly soluble in water; miscible with alcohol

Blood feeding behavior was studied by applying the repellents in various ways in the environment and on different chemosensory fields of female *Aedes aegypti*. Some general observations were also made on the blood feeding behavior of *Aedes* spp. mosquitoes in the field and *Aedes aegypti* in the laboratory. A preliminary test was made of the effect of washing chemosensory areas with lipoid solvents on blood feeding by *Aedes aegypti*.

REVIEW

Sense Organs of **Aedes aegypti** L.

An *Aedes aegypti* female is attracted to its host in part through the chemoreceptors located on head appendages, mainly the antennae. Bishop and Gilchrist (1946) showed that in *Aedes aegypti* eyes are not essential for feeding on blood. Roth (1951, p. 60) also reported that eyes are not necessary in locating the host in a small cage.

DeLong (1946) considered the antennae and the palps as the chief organs for locating the host and stimulating probing. According to him the antennae may perform both functions but the palps can receive stimuli only when the insect is directly on the skin. Roth (1951) considered that the antennae function as directional thermoreceptors and probably chemoreceptors as well. Roth (1951) also reported temperature receptors on the palps of *A. aegypti*. Dethier (1952) considered that

different receptor fields function at different levels of sensitivity. The antennae according to him are the most sensitive and the various mouth-parts less so. Rahm (1958) showed by antennal amputation that these organs are essential for host finding and attraction from a distance. He also reported that antenna-less mosquitoes can probe and suck if the palps remain intact.

Antennae

The antennae in the male and female consist of a basal ring-like scape, a globular pedicel, and a long flagellum of thirteen articles. The pedicel in both sexes contains Johnston's organ, which is more developed in the male.

Roth and Willis (1952) reported that many thin walled trichoid sensilla are present on each of the thirteen flagellar articles of the female *A. aegypti* and on the two terminal flagellar articles of the male. They concluded on experimental evidence that they serve as hygro-receptors.

Christophers (1960, p. 663) described the trichoid sensilla as "...40-50 μ in length, thin walled and without articulated base, arising from thin membrane over a pore canal surrounded distally by a semi-circular ridge in the article."

Steward and Atwood (1963) identified five structural types of sensilla on the antenna of the female *A. aegypti*. Three of these types they found thin walled and classified them as A1, A2 and A3. According to them a typical A1 sensillum is 0.06 mm long, curved and tapering to a sharp point. Type A2 is shorter, 0.04 mm long and with a blunt tip. Both are about the same diameter. The innervation of the two types is essentially the same. Steward and Atwood described type A3 as a short, curved, thin-walled peg organ which is innervated by a group of nerve cells. Sensilla of type A1 and A2 are more numerous on the distal articles while sensilla of type A3 are found to be located chiefly on the proximal articles of the antennal flagellum. They concluded from experimental evidence that type A1 and perhaps A3 play a major role in mediating attraction while type A2 are responsible for mediating repulsion.

Slifer and Sekhon (1962) studied the structure of the sense organs in the flagellum of *A. aegypti*. The heavy walled hairs according to them are mechanoreceptors. The thin walled hairs with sharp tips they thought to be chemoreceptors. The thin-walled hairs with blunt tips they supposed to be olfactory in function.

Palpi

Roth and Willis (1952) described the palps of female *Aedes aegypti* as abundantly supplied with thin-walled club-shaped sensilla on the terminal segment. Pointed trichoid sensilla are also present. There is also a central short sclerotized peg at the tip of the palp.

Labium

Frings and Hamrum (1950) noted four kinds of hairs on both sexes of *A. aegypti*. Of these, hairs about 40 μ long and lying at the tip of

the labella are considered to be tactile in function while curved hairs about 20 μ in length at the tip and on the ventral surface are believed to be chemoreceptors.

Tarsi
On the tarsi of the fore and mid legs of *A. aegypti* are many slightly curved hairs probably tactile in function (Frings and Hamrum, 1950). Wallis (1954) found that in *A. aegypti* all tarsal segments were provided with thin-walled curved spines. Slifer (1962) described the hairs on the tarsi as approximately 100 in number in the female. These hairs stain at the tip when dye is applied to the external surface of the insect. She concluded: "Little doubt now remains that the hairs with stainable tips are the tarsal gustatory receptors of the mosquito."

Mode of Action of Olfactory Receptors

Several theories have been advanced to explain the mode of action of olfactory receptors. Jones and Jones (1953) reviewed the modern theories on olfaction and classified them as; mechanical, chemical, steric, radiation and vibration theories.

Davies (1962) proposed that the mechanism of olfaction is the penetration and dislocation of a small region of the wall of an olfactory nerve cell. This dislocation allows the potassium and sodium ions to move across the membrane, so initiating the nerve impulse.

Amoore (1963), and Amoore, Johnston and Rubin (1964) favor the stereochemical theory of olfaction. According to them the odor of a chemical is determined by the structure of the molecule, in particular by its size and shape. If a chemical is volatile, and its molecules have the appropriate configurations to fit closely into the receptor site, then a nerve impulse will be initiated, possibly through a mechanism involving disorientation and hence depolarization of the receptor cell membrane.

Factors Attracting Mosquitoes to the Host

The mode of action of repellents cannot be fully studied without an understanding of the factors that attract the insect to the host. Contradictory views can be found in the literature on this point; all workers accept temperature and humidity, as attractant factors; others consider factors like carbon dioxide and host odor, or only carbon dioxide to be also important in attracting the mosquito to its host.

Howlett (1910) believed temperature to be the chief attractant and said that the smell of sweat or of blood was not attractive. Reuter (1936) showed that moisture was distinctly attractive to *A. aegypti*. Van Thiel (1937) assigned the role of attraction chiefly to the physical factors of temperature and humidity and the chemical factor, carbon dioxide. Later Van Thiel (1953) considered that the scent of the host plays an important part in the orientation of the mosquito toward it.

DeLong, Davidson, Peffly and Venard (1945) found moistened warm air more attractive to *A. aegypti* than warm air. Most of their tests were conducted with olfactometers or inanimate objects. Brown (1958) recognized six factors which guide female mosquitoes to their animal hosts, three of these being air-borne (water vapor, carbon dioxide, and

convective heat) and three visual (movement, contour, and reflectivity).

Kellogg and Wright (1957) and Wright (1962) considered moisture and carbon dioxide to be the main attractant factors. Christophers (1960, p. 535) remarked:"The evidence that smell is an important stimulus in the attraction of A. aegypti to feed is not very strong."

On the other hand, many have said that body odor plays an important role in the attraction of mosquitoes. Goeldi (1905) reported perspiration to be the agent attracting mosquitoes to man. Haddow (1942) reported that an unwashed African child attracts more Anopheles spp. than a clean child. Willis (1947) reported that females of A. aegypti and Anopheles quadrimaculatus Say were attracted by the odor of the human arm. He also found CO_2 in concentrations of 1, 10, or 50 per cent in the air not attractive to females of A. aegypti or Anopheles quadrimaculatus when tested in an olfactometer. Bates (1949) thought smell to be the primary stimulus in guiding the mosquito in its search for food. Rahm (1956) reported that CO_2 emitted by the skin did not determine attractiveness and remarked (1957) that human odor and sweat may play a part in the attraction of mosquitoes to the human hand. Again in 1957 he reported that perspiration did not seem to attract mosquitoes but the odors given out by the host did. Rahm (1958) further remarked that the olfactory substances of man were found to be alone responsible for greater activity of female A. aegypti. Dethier (1957) wrote:"Host finding and discrimination, trail following, orientation to odors by flying insects and courtship are shown to depend largely on the chemical stimuli."

EXPERIMENTAL - BEHAVIOUR

Blood Feeding in Relation to Repellents

Christophers (1960, p. 486) remarked on blood feeding by A. aegypti in the following words:"Another striking feature of feeding is that the insect once it has begun to suck blood, appears to become oblivious to all danger and considerable physical force is required to make it give up its hold." This feature is referred to by Gordon and Lumsden (1939) who wrote that they were only able to get A. aegypti to feed on the frog's foot by employing mosquitoes which had been allowed to start feeding on the human arm. When nearing repletion, however, the insect usually leaves readily if disturbed.

Kalmus and Hocking (1960) observed the effect of painting repellent with a fine camel hair brush on the backs of feeding mosquitoes. A lead was taken from this study and more observations were made on the effect of repellents on other species of Aedes in the field and Aedes aegypti in the laboratory.

Observations on Aedes *spp. in the Field*

For studies on the species of Aedes in the field a thicket of poplar trees was selected. The four repellents, dimethylphthalate, ethyl hexanediol, indalone, and N-N-diethylmetatoluamide were used. The mosquitoes reacted to all four repellents in the same way. The species of Aedes studied were A. punctor Kirby, A. cataphylla Dyar, and A. intrudens Dyar.

The time to take a complete blood meal, from the insertion of

the proboscis to its retraction after complete engorgement ranged from two to four minutes. (Mean = 2 min 31 sec with standard deviation 41 sec). It was observed that the mosquitoes could be very easily disturbed in the early stages of their blood meal. If a clean brush were brought near them soon after the insertion of the proboscis, they could be seen retracting it. If a repellent or olive oil were placed near the antennae or painted on the mesonotum, the mosquitoes invariably flew away. As reported by Kalmus and Hocking (1960, p. 7) "A contact between repellent chemicals as liquids and substantial areas of the proboscis, tarsi and tibiae, mesonotum or the wings leads to the interruption of biting, and in mosquitoes not engaged in biting to the retraction of the touched limb or limbs or to take off." But the behavior of mosquitoes was found quite different in relation to repellents and other stimuli if they had been feeding for a minute or more, i.e. roughly in the middle of their meal; e.g.:

(i) The mesonotum was rubbed with a dry brush, painted with repellents or olive oil until the whole mesonotum was covered with liquid, but the mosquito never flew away, instead it completed its blood meal, continuing to feed for another 45 seconds to one minute.

(ii) The antennae were painted with repellents, were in fact soaked in repellent, but the mosquitoes continued to feed.

(iii) A drop of repellent was made to flow near the tarsi, there was no reaction until it made contact with them. As soon as contact was made the tarsus was lifted. The same reaction was observed with olive oil. However, the mosquitoes continued to feed even when the tarsi of all the six legs were lifted. The mosquito then came to rest on its abdomen. When the repellent was presented on a brush near the lifted tarsi, they sometimes rested the tarsi on the repellent soaked brush, without showing any other abnormal behavior, and continued to feed.

(iv) Similar behavior was observed in mosquitoes feeding on the foot through socks. Mosquitoes coming to feed landed only on clean areas of the sock and avoided areas where repellent had been placed. However, mosquitoes which had been feeding through the sock for some time were not affected if a repellent was placed on the sock underneath them, and they continued to feed to completion although they lifted the abdomen.

(v) Chloroform or ether was brought near the abdomen of a feeding mosquito. It always flew away, even when it had been feeding for a minute or more.

(vi) A hot spatula was brought near the mosquito (about 1 mm). The spatula was heated for two minutes in a flame of a spirit lamp. Eighty per cent of the mosquitoes took off in 5 to 10 seconds. When the spatula heated for the same time was kept at the same distance from the mercury bulb of a Fahrenheit thermometer, the thermometer registered a rise of 4-6 degrees.

(vii) Repellent was painted on the wing of a feeding mosquito. The mosquito always flew away but when the wing was rubbed with a dry brush or painted with olive oil it continued to feed.

(viii) Physical injury was inflicted on the mosquito to the extent

that all the six legs were clipped off at the femoro - tibial joint, but it continued to feed and did not fly away.

The observations were made at a temperature of 65°F and R. H. of 57%.

Observations on Aedes aegypti

In the laboratory the same behavior was studied in *Aedes aegypti*. A one cubic foot cage made of steel wire and covered with nylon net was fitted with a sleeve on each of two adjacent walls, i. e. at right angles to one another. Mosquitoes were allowed to feed on a hand inserted through one sleeve while the other hand was introduced through the other sleeve to apply the repellent.

As observed in the other species of *Aedes*, *Aedes aegypti* could also be easily disturbed in the initial stages of blood feeding, but after one minute of feeding they could not be disturbed so easily:

(i) When the mesonotum was rubbed with a dry brush or painted with olive oil or any of the four repellents under study.

(ii) When their wings were painted with repellents. This was contrary to the behavior observed in the field species which invariably flew away whenever repellents were painted on the wings.

(iii) They continued to feed even when they were made to rest their tarsi on the repellent soaked brush.

(iv) Being small in size, it was not possible to paint their antennae with repellent while they were feeding, but when a drop of repellent was placed very close to the proboscis they continued to feed.

(v) Almost every mosquito continued to feed when the tarsi of its hind legs were clipped off, but some flew away when the tarsi of their other legs were clipped.

(vi) When a heated spatula was brought near them they always flew away even when the spatula was as far as 1-2 cm away. It had to be brought much nearer to mosquitoes in the field to elicit this response. When the spatula heated for the same time was kept at the same distance from the mercury bulb of a Fahrenheit thermometer this registered a rise of 1.5 to 2 degrees.

Experiments were conducted by applying the repellent on different chemosensory fields of female *A. aegypti* and observing the behavior and recording the number feeding on an untreated human arm. As the repellent was not applied on the skin, there was no interaction between the skin and the repellent or the chemical stimuli emanating from the skin and the repellent on the surface of the skin. The experiments provided some understanding of the site of action of different repellents as well as providing a quantitative basis for comparing the repellents with each other. The experiments also provided a quantitative basis for evaluating the function and efficiency with which the different chemosensory fields play their role in the act of feeding as well as some grounds for accepting the role of smell in attracting mosquitoes to feed and the function of the repellent when applied on the skin in offsetting this role.

10-12 female mosquitoes, 7-8 days old, previously fed on raisins and sugar solution only, in a sucking tube and then chilling them for 1.5 min at 15°F, in order to immobilize them. Their probosicides, either one or both antennae, or all the tarsi, were then painted with repellents with a fine brush in separate sets of experiments. This operation was performed over a cold petri dish covered with a filter paper and placed under a binocular microscope. A radius was drawn in ink on the filter paper and mosquitoes were treated one by one, starting on one side of the radius until all of them were treated. They were then sucked back into the sucking tube and released in a paper lined petri dish to revive in a one cubic foot cage of steel wire covered with nylon net. The mos - quitoes recovered from the chill in 2-3 minutes. The behavior and the number that fed on blood on introducing the arm into the cage through a sleeve were noted, firstly ten minutes after the treatment and then at greater intervals from the treatment until the number fed in a given time approached the number fed in controls. Two controls were run with each set of experiments, one a plain control when the receptor field that was intended to be treated was rubbed with a dry brush only, and another when it was painted with olive oil. The palps could not be treated separately without running some repellent on the proboscis and the antennae, because of their close proximity to these structures.

Results - The figures given in table 2 give the cumulative mean percentages of mosquitoes feeding on blood after different chemoreceptor sites were painted with repellents. The standard error of the mean was used to find statistical significance between the means.

The results show that Rutger's 612, diethyl toluamide, and indalone reduce the number of mosquitoes feeding on blood more than dimethyl phthalate after the first ten minutes when the proboscis was painted, and the effect lasted longer. Indalone remained significantly more effective as compared to Rutger's 612 and diethyl toluamide after two hours when painted on the proboscis.

When painted on both the antennae, diethyl toluamide, Rutger's 612 and indalone again reduced the number of mosquitoes feeding more than dimethyl phthalate. The effect of dimethyl phthalate was found to have been lost within one hour but the effect of the other three repellents lasted more than six hours.

When painted on one antenna, the same significant differences were found between the repellents as when both the antennae were painted, i.e., diethyl toluamide, Rutger's 612 and indalone were significantly more effective than dimethyl phthalate.

The results obtained on painting all the tarsi with repellents were, however, different. Dimethyl phthalate was found to reduce feeding more effectively when painted on tarsi than when painted on both the antennae or on the proboscis, and to maintain this effect at least as long as the other three materials.

There is evidence that many repellents work by way of specialized chemoreceptors (Weismann and Lotmar, 1949; Dethier and Yost, 1952; Peters, 1956; Dethier, 1956 a). Peters (1956) reported that *Calliphora erythrocephala* could detect dimethyl benzamide with the tarsal

TABLE 2 - Cumulative percentages of *A. aegypti* females that fed after different chemoreceptor areas were painted with repellents. Means of four replicates ± standard errors.

Chemoreceptor area painted	Observation time	Control	Olive Oil	D.M.P.	D.E.T.	Rutger's 612	Indalone
Proboscis	10 min	82 ± 3.1	73 ± 2.5	33 ± 3.6	12 ± 3.9	22 ± 5.5	10 ± 3.5
	1 hr	- -	- -	72 ± 4.4	51 ± 8.7	52 ± 3.7	47 ± 3.2
	2 hr	- -	- -	75 ± 4.7	72 ± 2.9	76 ± 5.7	63 ± 2.6
	3 hr	- -	- -	- -	- -	- -	71 ± 1.6
Both Antennae	10 min	76 ± 2.3	71 ± 4.1	33 ± 2.3	3 ± 3	0	3 ± 3
	1 hr	- -	76 ± 2.5	81 ± 4.2	6 ± 3.2	5 ± 2.8	8 ± 4.8
	2 hr	- -	- -	83 ± 2.3	11 ± 4.1	11 ± 4.1	19 ± 5
	6 hr	- -	- -	- -	32 ± 3.1	33 ± 4.5	30 ± 3.1
One Antenna	10 min	85 ± 2.9	80 ± 4	61 ± 3.3	5 ± 2.7	25 ± 5.9	19 ± 3.3
	1 hr	- -	- -	80 ± 4	38 ± 10	49 ± 4.3	56 ± 2.4
	2 hr	- -	- -	- -	75 ± 2.3	69 ± 3.2	72 ± 4.5
Tarsi	10 min	80 ± 4	73 ± 2.5	27 ± 2.7	14 ± 5.2	27 ± 2.9	30 ± 3.8
	1 hr	83 ± 2.5	78 ± 2.5	50 ± 8.4	61 ± 2.3	71 ± 3.9	74 ± 3.5
	2 hr	- -	- -	68 ± 1.5	76 ± 2.5	76 ± 2.2	79 ± 1.5

receptors only, while other materials like indalone and dimethyl carbate could be detected with the tarsal receptors, labella, and antennae.

The significant difference in the number of mosquitoes landing on the hand after treatment of chemoreceptors on different head appendages and on the tarsi can be explained on the basis of the population of chemoreceptors getting such treatment. As most of the chemoreceptors are situated on the antennae, their treatment with repellents would inhibit the landing of mosquitoes on the hand more than the treatment of other head appendages. The ineffectiveness of the painting of one antennae only in keeping the mosquitoes from a blood meal for two hours can be explained by the same argument, i.e., a large population of chemoreceptors remained functioning effectively when only one antenna was painted. The painting of any one of these chemoreceptor sites with repellent must be affecting the mosquito in two ways, affecting the chemoreceptors of the chemosensory area painted in liquid form and also affecting the adjacent chemosensory sensilla in vapor form. The greater the area painted, the greater the number of sensilla affected, resulting in inhibition of feeding for a longer period.

Painting Repellent on Mosquito Antennae and Host Skin

By the procedure described above one antenna of each of about 10 *A. aegypti* females was painted with diethyl toluamide. An arm also treated with diethyl toluamide was then introduced into the cage and the behavior of the mosquitoes was studied. A little more flight activity and some searching on the wing was observed in these mosquitoes as compared to those in the control where no repellent was applied on the mosquitoes themselves but only on the hand. A similar behavior was observed in experiments with the other three repellents as well. In controls, mosquitoes were seen mostly sitting on the walls of the cage. There was little or no flight activity.

When both the antennae of mosquitoes were treated with diethyl toluamide, indalone or Rutger's 612, and the same repellent was applied on the hand introduced into the cage, the mosquitoes could be seen searching on the wing. Many landed on the repellent coated surface of the hand, walked about and even probed but did not take a blood meal. The behavior was observed for ten minutes every hour for four hours but no mosquito bit. In similar experiments with dimethyl phthalate, however, no landings on the hand were observed though the mosquitoes came quite close to it and sometimes even touched the skin.

Sugar Feeding

The principal food of female *Aedes aegypti* is blood from a human host though they can exist for long periods on food other than blood. Male *Aedes aegypti* do not take blood at all but feed entirely on sugary materials. Goeldi (1905) kept females alive for 31 to 102 days on honey alone. Macfie (1915) observed that the females feed on honey for the first couple of days but the males feed only on honey at anytime. Gordon (1922 b) observed both males and females of *Aedes aegypti* sucking nectar from flowers. Many observers have noted that sugary fluids, raisins, bananas, and other fruits are sucked by both sexes.

Many workers have devoted much time to studies of the effect of repellents on blood feeding of mosquitoes but their effect on sugar feeding has not attracted much attention. Evans (1961) has studied the effects by the blowfly *Phormia regina* Meigen. Experiments were conducted to study the effect of repellents on the feeding of *Aedes aegypti* on raisins.

Kalmus and Hocking (1960) conducted some tests on blood feeding in relation to repellents with *Aedes aegypti* by keeping a 10 cm length of 3mm outside diameter glass tubing which was clamped in a vertical position so that the lower end was about 1 cm above the middle of a 6 cm bare circle on the back of a gloved hand. A few drops of repellent were placed in the lower end of the tube. In this way a circle of skin about 1.5 cm diameter was kept free of bites. A lead was taken from this experiment in exploring the effect of repellents on the feeding of *Aedes aegypti* on raisins.

Experiment 1

About 100 male and 100 female mosquitoes were taken in a cubic foot cage of steel wire covered with nylon net. The age of the mosquitoes was 2-4 days and they were not fed anything for six hours prior to experiments. Ten raisins were fixed with 1 cm clear space between each on a horizontal steel wire hanging 4 inches below the top of the cage. The wire was hung by bending its ends and hooking them on top of the side walls of the cage. A 2 cm wide strip of paper was fixed above the raisins, running parallel to them at a distance of 1.5 cm. Half of this paper strip (covering 5 raisins) was painted with repellent and the other half (covering the other 5 raisins) was kept as a control. Observations were made on the number of mosquitoes settling on either side at intervals of 5 minutes. After each observation the cage was shaken and another observation recorded after five minutes. In this way five replicates were taken for each repellent. Separate batches of mosquitoes were taken in separate cages for experiments with different repellents.

The observations are recorded in table 3. The vapor of repellents significantly reduced the number of mosquitoes feeding on raisins. The standard error of the mean was used as statistical test for significance.

Experiment 2

Ten raisins were fixed on the wire lying as close to each other as possible without touching. Five alternate raisins were then painted with repellent leaving the other five as controls. The numbers of mosquitoes that settled on the treated and untreated raisins are recorded in table 4, column 1 to 3. The figures are means of 5 replicates. Observations were recorded every five minutes as in the previous experiment. The total number of mosquitoes in the cage for each experiment was 200.

In these experiments mosquitoes were seen coming close to the raisins to land but they usually flew away without landing. No significant difference was found between the number of mosquitoes settling on treated

and untreated raisins in the control with olive oil. The results show
that the repellent on the treated raisins kept the mosquitoes away from
the untreated raisins as well. Kalmus and Hocking (1960, p. 23) obtained
bites up to almost a mosquito half-width (about 2.3 mm) from a repellent
painted circle on the back of the hand. In these experiments the mean
width of untreated raisin separating the two treated ones with repellent
was 10 ± 0.3 mm. This greater distance was perhaps due to the factors
of heat, CO_2 and probably skin odor, which were missing as attractant
factors in the raisins.

TABLE 3 - Numbers of A. aegypti settling on raisins separated by 1 cm,
under the plain and repellent coated halves of a paper strip.
Means of five replicates ± standard errors.

Paper strip half	Olive oil	D.M.P.	D.E.T.	Rutger's 612	Indalone
Painted with chemical	18 ± 1.9	5 ± 0.5	3 ± 0.7	3 ± 0.1	2 ± 0.8
Plain	19 ± 4	18 ± 1.9	12 ± 1.3	13 ± 1.4	10 ± 2.2

Experiment 3

Raisins were kept 1 cm clear apart from each other and alternate
raisins were painted with repellent. Other factors were the same as in
the previous experiments.

The mean numbers of mosquitoes that landed on the treated and
untreated raisins are given in table 4, columns 4 and 5. The numbers
of mosquitoes feeding or settling on the untreated raisins were still very
low and no significant difference was found in the number of mosquitoes
feeding on untreated raisins in this experiment as compared to the number
of mosquitoes feeding on untreated raisins in the previous experiment.

Experiment 4

Only 5 raisins were taken and were placed 1 cm clear apart and
the portion of wire between them was painted with repellent. Since the
raisins were not painted with repellent in this experiment their number
was reduced to five so that the number of mosquitoes landing on them
could be compared with the number of mosquitoes landing on the untreated
raisins in previous experiments.

The results are given in table 4, column 6. The comparison of
results in table 4 shows that significantly more mosquitoes settled on
raisins in this experiment than in experiments where treated and
untreated raisins were placed close to each other. This is perhaps due
to the small surface area of wire between the raisins as compared to
the much greater area of the raisins in the previous experiments. This

would result in a much slower production of repellent vapor.

TABLE 4 - Numbers of *A. aegypti* settling on raisins in the presence of repellents. Means of five replicates ± standard errors.

	Raisins close together, alternate raisins painted with repellent		Raisins 1 cm apart, alternate raisins painted with repellent		Raisins 1cm apart & the wire in between painted with repellent
Chemical	Untreated raisins	Treated raisins	Untreated raisins	Treated raisins	Untreated raisins
Olive oil	16 ± 2	14 ± 4	10 ± 0.7	11 ± 1.4	10 ± 1
D.M.P.	1	0	2 ± 0.5	0	2 ± 1
Rutger's 612	1	0	2 ± 0.8	0	3 ± 0.5
Indalone	0	0	0	0	3 ± 1
D.E.T.	0	0	2 ± 0.5	0	5 ± 0.5

Mating

    In *Aedes aegypti* "The stimulus which induces the male to copulate is the sound produced by the female during flight." ".. odor plays no part in the sexual behavior of *aegypti* ..." (Roth, 1948, pp. 284, 282). Roth also observed that in *Aedes aegypti* the male is the aggressor and is attracted by the female in flight and that the female is passive and does not show any mating behavior similar to that of the male. "...never in our observations was a male seen to initiate copulation with a resting female" (Roth, 1948, p. 276). Banks (1908, p. 246) on the contrary stated that specimens of *aegypti* confined in small jars"...have been seen to copulate while the female hangs from the gauze covering the vessel, the male always approaching her from the ventral surface." Christophers (1960, p. 502) observed that copulation takes place quite commonly with the female at rest.

    During the course of this work it was observed that a female *Aedes aegypti* is not entirely passive and that copulation does take place when a female is at rest. It was observed that when a flying male came close to a sitting female, the female would take flight and the male would grasp her for copulation. Many times females were seen taking flight spontaneously and males were seen getting hold of them in mid-air. The males were also observed coming to land sideways with a female,

then trying to take a ventral position and many a time they succeeded. At other times because of his efforts to gain a ventral position to the resting female the male roused the female to fly and copulation took place on the wing or the two could be seen falling to the floor copulating. But mostly copulation took place with a female in flight.

Roth (1948) also observed that the male would copulate repeatedly with the same or different females. After repeated matings, females become more and more reluctant to fly and would resist the attempts of the males to copulate. Richards (1927) suggested that repeated copulations exhaust the individuals. Shannon and Putnam (1934) in their laboratory study of *A. aegypti* observed that the average pupal period of females was 14 hours longer than that of males. Roth (1948, p. 308) observed that by the time the female begins to fly and becomes 'attractive' the male's antennae have reached a state where the sound stimulus can be perceived and his genitalia have rotated sufficiently so that copulation can be successful (usually about 15 to 24 hours after emergence). In view of these observations it was necessary in this work to separate the sexes before they started mating and to keep ·the observation time reasonably short. To forestall fatigue in the females due to repeated copulations, the males were separated from the females 14 hours after emergence.

Ten females 2-4 days old and 10 males 5-6 days old were used for each experiment. The females were chilled in a sucking tube for 1.5 minutes at 15°F and then all their tarsi were painted with repellent with a fine brush while on a cold petri dish under a binocular microscope. Since *Aedes aegypti* mate venter to venter and the female does not clasp the male to her, her legs remaining out-stretched and serving as structures to which the male clings (Roth, 1948, pp. 270, 301), it was decided to paint the tarsi of the female mosquito with repellent. After the tarsi were painted the females were released in a one cubic foot cage and allowed to recover from chill. They recovered in 3-4 minutes. Ten minutes after the treatment 10 males were released in the cage. After application of the repellent on the tarsi of the female *Aedes aegypti* few flew spontaneously. Most females sat quietly on the walls of the cage. Males hardly ever succeeded in persuading the female at rest to copulate. It was also observed, though no quantitative basis could be laid down for this, that the efforts of the male to copulate with the resting female, as well as with the female in flight, were less persistent and quite often they were seen releasing the female soon after coming in contact. The cage was therefore shaken every minute to make the females fly and the number of matings in a period of 30 minutes was recorded. Each experiment was performed with a new batch of mosquitoes.

The results are recorded in table 5. The standard error of the mean was used as a test of significance. The highly significant reduction in the number of matings in *A. aegypti* in association with repellents can be explained as a result of two factors: 1) a decrease in the flying activity of the females and 2) less persistent efforts by males and premature release of the female.

Though the cage was shaken every minute in experiments with repellents as well as in the control it was observed that the females in

the controls continued to fly for a much longer time after shaking than in experiments with repellents. With repellents, most of the time the females could be seen coming to rest on the wall immediately after shaking the cage, and many a time on shaking they would fly only from one wall of the cage to another. There was also a lack of spontaneous flight activity on the part of the females.

TABLE 5 - Numbers of matings in a 30 minute period in a population of 10 male and 10 female *Aedes aegypti* with repellents applied to the tarsi of the females. Means of four replicates ± standard errors.

Control	Olive oil	D. M. P.	Indalone	Rutger's 612	D. E. T.
65 ± 2	65 ± 1	33 ± 2	30 ± 3	33 ± 3	33 ± 3

Oviposition

Wallis (1954) in his studies on the oviposition activity of mosquitoes, including *A. aegypti*, found that the female could detect an objectionable amount of salt even when the movements of the abdomen were restricted. Likewise surgical removal of the palpi, proboscis, and antennae from the head did not result in loss of sensitivity. Surgical removal or wax coating of various combinations of legs and leg articles resulted in the demonstration that sensitivity was localized in the tarsal articles of all the species of mosquitoes studied by him. His investigations also showed that the sensitivity was present in all the tarsal articles of *Aedes aegypti*. The thin walled chemoreceptors of the tarsi enabled the mosquitoes to detect differences in saline concentrations as slight as 0. 02 M.

Browne (1960) studied the role of olfaction in the stimulation of oviposition in the blowfly *Phormia regina* Meigen. He found that the odor of a liquid medium containing powdered milk and yeast stimulated the blowfly to oviposit. He also provided evidence for olfactory perception by the ovipositor of the blowfly.

In this study oviposition in *Aedes aegypti* was observed by associating potential oviposition sites with repellent vapors as well as by applying repellents on the tarsal chemoreceptors.

Experiment 1

Five, 7-8 day old blood fed females in a one cubic foot cage were taken for each experiment. The cage was provided with a rectangular platform, 7"x 4" made of a steel wire frame (diameter of wire 2.5 mm). The platform was covered with nylon net on one side and with two paper towel strips pasted on the other except in the center where a gap of 1

cm was left in between the strips, see figure 1.

The platform was placed in the cage, nylon net side upwards, the ends resting on two glass bottles filled with water. On the nylon net was spread a piece of cheese cloth, the two ends of which remained dipped in the water in the glass bottles. The cheese cloth was kept wet by capillary action by the water in the two bottles. One of the two paper strips was painted with repellent while the other was left untreated. Thus an oviposition platform for the mosquitoes was provided, one half of which had repellent vapor coming from underneath through the nylon screen, while the other half served as control. The nylon net underneath the cheese cloth served as a support for it and did not allow it to come in contact with the repellent on the paper strip below but allowed the repellent vapors to pass through. Most of the eggs were found to be laid on the cheese cloth but some were laid on the paper strip. They were counted separately 72 hours after the blood meal, and the results are recorded in table 6. Four experiments were run with each repellent.

The behavior of *Aedes aegypti* during egg laying is described in detail by Wallis (1954). During the experiments it was observed that a female mosquito could sample the oviposition sites while on the wing and would land on the control half rather than on the repellent treated half of the oviposition platform. At other times when she landed on the repellent half she walked for a few seconds and then flew away and landed on the control side. This behavior demonstrates the function of olfactory receptors in the selection of an oviposition site when repellent vapors are associated with it. The complete absence of egg laying on the repellent coated as well as olive oil coated paper towels on the lower side of the platform seems to be the result of tarsal chemoreceptors which select the suitability of the egg laying medium on contact. The significantly small numbers of eggs laid on cheese cloth on the repellent side as compared to the number of eggs laid on the control side show that *Aedes aegypti* rejects oviposition sites when these are associated with repellent vapor.

Experiment 2

Experiments were also conducted by painting the tarsi with repellent by the same technique as described in previous experiments and recording the number of eggs laid in 24 hours. Christophers (1960, p. 507) records that egg laying in *Aedes aegypti* usually begins on the afternoon of the third day from blood feeding, counting the day of feed as zero. Female mosquitoes 6-7 days old were fed on blood and left in a cage with raisins for three days. On the fourth day their tarsi were painted with repellent and the mosquitoes were placed singly in separate vials with water soaked cotton wool in the bottom and a nylon net cap on the top on which was placed a raisin. Eggs laid in a 24 hour period were then counted. Four replicates were run for each experiment. The mean numbers of eggs laid are recorded in table 7.

The difference in the number of eggs laid in the control and those laid by repellent treated mosquitoes is not significant, using standard error of the mean as a test of significance. This is perhaps
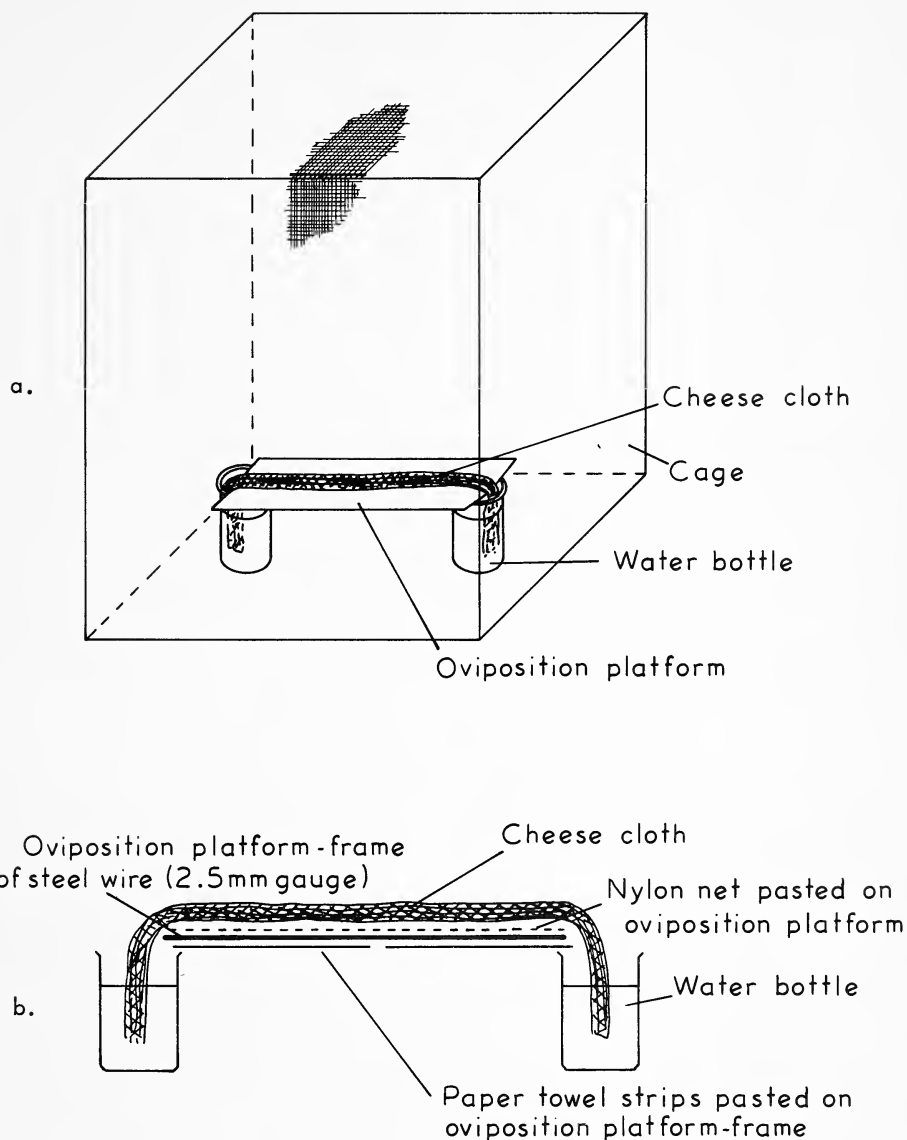
Figure 1. Diagrams showing: (a) arrangement of the oviposition platform in the cage, and (b) a vertical section of the oviposition platform.

due to the fact that the mosquitoes had no opportunity to select a site for oviposition as they were confined in small vials.

TABLE 6 - Numbers of eggs laid by *A. aegypti* females in the presence of repellents. Means of four replicates ± standard errors.

Chemical	Eggs laid on control side		Eggs laid on repellent side	
	On cheese cloth	On paper towel	On cheese cloth	On paper towel
Rutger's 612	128 ± 18.6	52 ± 4.2	0	0
D.E.T.	160 ± 20.5	47 ± 2.2	2 ± 1.7	0
Indalone	156 ± 18.4	19 ± 1.5	0	0
D.M.P.	208 ± 13	22 ± 1.7	5 ± 1.1	0
Olive oil	87 ± 6	12 ± 1.1	68 ± 7.2	0

Experiment 3

Experiments were also conducted to determine whether antennectomized mosquitoes would discriminate between the control and the repellent sides of the oviposition site. Twenty female mosquitoes which had been fed on blood previously were operated upon for each experiment on a cold petri dish under a binocular microscope after first chilling them for 1.5 minutes at 14°F. Ten to 12 flagellar segments of the antennae were excised and the mosquitoes then released in the cage with the oviposition platform shown in figure 1.

Though sometimes mosquitoes could be seen sitting on the control side of the egg laying platform, no eggs were laid in any of the experiments over a week's time except in the experiment with diethyl toluamide where there were 4 eggs on the control side. A high mortality (70-75%) was also observed in mosquitoes during this period. The almost complete absence of oviposition by antennectomized mosquitoes may be due to lack of orientation of mosquitoes to the water soaked cheese cloth on account of the great reduction in the number of hygroreceptors as a results of excision and consequently a great increase in the threshold of moisture perception. The high mortality rate can also be assigned to the same factor, i.e., lack of orientation to the water soaked cheese cloth and hence dehydration. Mosquitoes were seldom seen sitting on the wet cheese cloth. Most of the time they were found sitting on the walls of the cage with very little flight activity. The very low activity in antennectomized mosquitoes confirms the findings of Bar-Zeev (1960) who found only 4 per cent of mosquitoes could be activated when antennectomized as compared to 60.1 per cent when intact.

TABLE 7 - Numbers of eggs laid by single *A. aegypti* females after painting the tarsi with repellents. Means of four replicates ± standard errors.

Control	Olive oil	D. M. P.	Rutger's 612	D. E. T.	Indalone
39 ± 8.8	42 ± 4.8	29 ± 5.2	34 ± 8.6	34 ± 5	33 ± 4.4

Experiment 4

Experiments were also conducted to test oviposition after treating the terminalia of the females with repellent. The female *aegypti* mosquitoes were fed on blood when 7-8 days old, and their terminalia painted with repellent 72 hours after the blood feed by the same technique as described in the previous experiments, and then released in the cage.

All the mosquitoes became too crippled to move about or fly shortly after the painting of the tip of the abdomen and died in a few hours.

Behaviour in Relation to Wind Direction and Speed

Kalmus and Hocking (1960, p. 21) conducted a series of experiments in which target areas were drawn out on the backs of subjects who wore shirts with the backs cut out. They recorded the distribution of bites in relation to a small repellent treated area. To demonstrate the effect of wind direction on the distribution of bites in relation to repellent, experiments were conducted in the laboratory on *Aedes aegypti* using the same technique.

Experiment 1

A circle of 3.5 cm radius was drawn in hard clear nail varnish on the bare chest of a subject. Concentric to this another circle of 6.5 cm radius was drawn. The outer circle was divided into two equal halves by drawing a diameter. A hair drier was used to produce the air current and a variable transformer was included in the circuit to permit adjustment of the speed of the wind. The wind speed was kept at 43 cm/sec and its direction at right angles to the drawn diameter. The source of wind, i.e., the nozzle of the blower was kept 23 cm away from the central circle which was coated with repellent. The blower was kept in such a position as to give a uniform flow of air over the marked area. The repellent used was diethyl toluamide. One hundred 7-8 day old female *Aedes aegypti* mosquitoes were taken in a one cubic foot cage of steel wire with nylon net around it for each experiment. The mosquitoes were fed on sugar solution only before the experiment. The cage was placed on the marked area and the portion of skin outside the marked area was covered with a polyethylene sheet. Mosquitoes soon started biting through the nylon net on the floor of the cage. An observer kept a record of the mosquitoes that settled and flew away, or settled and bit, in the upwind and downwind halves of the circle. The counts

were made for 5 minutes in each experiment. Controls were run with the same wind speed without repellent. The number of mosquitoes that settled or bit in the upwind and downwind halves of the circle are given in table 8.

The results show that in the control where repellent was not painted in the central circle, significantly more mosquitoes settled or bit on the downwind side of the circle than on the upwind side. This is in conformity with the observations made by Kalmus and Hocking (1960, p. 4) with field mosquitoes. However, when repellent was painted in the central circle it was observed that the number of mosquitoes settling or biting on the downwind half of the outer circle was significantly lower than the number settling or biting on the upwind half. This was due to the presence of repellent vapor carried by the wind on the downwind half of the outer circle.

TABLE 8 - Numbers of *A. aegypti* females settling or biting in relation to wind direction and D. E. T. on the marked area of skin.

Wind speed	Control*		D. E. T. **	
	Upwind	Downwind	Upwind	Downwind
43 cm/sec	15 ± 2.5	27 ± 1.6	35 ± 2.9	8 ± 1.7

*   Means of two counts ± standard error
** Means of three counts ± standard error

Experiment 2

In another set of experiments the effect of different wind speeds was determined on the settling and biting of mosquitoes in relation to repellent. Experiments were conducted in a similar fashion as described under the experiments with different wind directions, except that the portion of the body used was the thigh instead of the chest, which gave the advantage of the subject himself making notes of the number of mosquitoes landing or biting. A control was run with each wind speed and all the controls with different wind speeds were run first in order to avoid contamination of skin area with repellent vapors. After the controls were run, different batches of mosquitoes were then used in experiments with the same wind speeds in relation to repellent painted in the central circle. The repellent used was diethyl toluamide. The portion of skin outside the outer circle was covered with polyethylene sheet and the count of mosquitoes settling or biting in the upwind or downwind half of the circle was recorded for five minutes in each experiment.

The results are shown in table 9. In previous experiments with different wind directions the number of mosquitoes settling or biting in the upwind half of the circle in experiments with repellents was

significantly higher than the number of mosquitoes in the downwind half of the circle. The results given in table 9 show that the mosquitoes continue to show the strong tendency of settling more on the upwind side in relation to repellent with different wind speeds.

The maximum wind speed at which mosquitoes were able to settle on a bluff body was reported to be 95 cm/sec and that of settling on the streamlined body to be 55 cm/sec. Kalmus and Hocking (1960, p. 15). In this case the maximum speed of wind at which the mosquitoes settled on the skin was 265 cm/sec which is very high as compared to the wind speed with the bluff or streamlined bodies. This is probably due to the attractant factors of the skin acting on the mosquitoes.

TABLE 9 - The number of *A. aegypti* females settling or biting in the up-
wind or the downwind half of the circle marked on skin in
relation to different wind speeds and diethyl toluamide.

Wind speed	Control		D. E. T.	
	Upwind	Downwind	Upwind	Downwind
0 cm/sec	27	29	23	19
43 cm/sec	13	26	41	12
134 cm/sec	16	31	9	4
190 cm/sec	5	14	3	1
227 cm/sec	4	8	5	0
265 cm/sec	4	6	2	0
314 cm/sec	0	0	0	0

Orientation to Gravity and Centrifugal Force

Experiment 1

To study the orientation of *Aedes aegypti* to gravity in relation to repellents, experiments were conducted in a plastic petri dish of 9 cm diameter. The lid of the petri dish was perforated with 2 mm diameter holes, about 9 holes per sq cm to allow the repellent vapors inside the dish to escape. The floor of the petri dish was lined with a filter paper which was divided into four quadrants designated top, left, bottom, and right.

Twenty female mosquitoes, 7-8 days old were taken, chilled for 1.5 minutes at 14°F and then released in the petri dish. On recovery

of mosquitoes from chill the petri dish was turned with a diameter vertical and given five complete turns on the horizontal axis through its center; thereafter the position and the number of mosquitoes was noted in each quadrant after a minute. The experiment was replicated five times without repellent as a control. A band of repellent 1 cm wide was then painted on the outer margin of the top quadrant. Mosquitoes were chilled and placed in the petri dish and allowed to recover. After the mosquitoes had completely recovered, the dish was given five complete rotations as in the control, keeping it vertical and rotating it about its horizontal axis. The experiment was repeated five times with each repellent.

In the control the mosquitoes could be seen walking upwards and most of them collected in the top quadrant. Significantly less mosquitoes remained in other quadrants. Almost all the mosquitoes were seen facing upwards and the root mean square deviation of their body axes from the vertical axis of the petri dish was found to be zero.

With repellent significantly less mosquitoes entered the top quadrant. Most of them remained in the left, right, and bottom quadrants. They were also seen walking at an angle to the repellent or turning away from it. Their angle of turning (i.e., the angles which the longitudinal axes of the bodies formed with the vertical axis of the petri dish) was noted by marking their position in each quadrant on a separate sheet of paper and then measuring the angle and direction of inclination to the vertical.

Table 10 shows the distribution of mosquitoes in the various quadrants of the petri dish in the presence of repellents, and table 11 shows the root mean square of the angle of inclination of the body axes of mosquitoes to the vertical in the presence of repellents in the petri dish.

Results with olive oil were not found to be significantly different from those of the plain control.

The effect of the presence of repellent on the head upwards orientation of the mosquitoes in relation to gravity was highly significant.

Experiment 2

The effect on geotaxis of painting repellent on the mesonotum and the antennae was also observed. Seven to 9 days old female mosquitoes were chilled for 1.5 minutes at 14°F and their mesonota or antennae were painted with repellent. They were then placed in a 9 cm petri dish having holes in the lid and lined with filter paper. After complete recovery of the mosquitoes from chill the dish was held with its central axis horizontal and rotated slowly about this, one rotation in 20 seconds, and the positions of the mosquitoes were noted. Normal female *A. aegypti* show a counter rotation to maintain a head upward under these circumstances (Kalmus and Hocking, 1960, p. 8).

The mosquitoes with their mesonota painted oriented facing upwards by counter rotation but when the antennae were painted with repellent, on placing the dish in a vertical position the mosquitoes could be seen sitting on the vertical surface head upwards cleaning their antennae with the tarsi of the forelegs. When the dish was rotated slowly

while they were cleaning their antennae, they did not react until they faced downwards. Then they were found to lose their balance and were seen to place their forelegs on the vertical surface. Some of them turned around, faced upwards and started cleaning the antennae again, but typical counter rotation was absent.

TABLE 10 - Numbers of *A. aegypti* females found in different quadrants in relation to repellents. Means of five replicates ± standard errors.

Chemical	Top	Left	Bottom	Right
		Quadrants		
Control	15 ± 1	2 ± 0.5	1 ± 0.4	2 ± 1
Olive oil	13 ± 1.6	3 ± 0.8	2 ± 0.5	2 ± 0.7
D.M.P.	3 ± 0.4	4 ± 1	4 ± 1	9 ± 1.3
D.E.T.	2 ± 0.1	6 ± 0.8	6 ± 1.4	6 ± 0.8
Indalone	4 ± 1	5 ± 1.2	6 ± 1.4	5 ± 1.3
Rutger's 612	2 ± 0.5	5 ± 0.4	8 ± 1.3	5 ± 1

Experiment 3

According to Kalmus and Hocking (1960, p. 8), when mosquitoes were centrifuged in a 9 cm petri dish at 390 rpm and observed under stroboscopic illumination, they were found facing towards the center of the dish, and sometimes walking towards it.

In this study of the same behavior in relation to repellents a plastic petri dish of 9 cm diameter was lined with filter paper on which one radius was drawn in ink. Its lid was extensively perforated by small holes. Mosquitoes, both males and females (50 to 60 adults) were released in this dish and centrifuged at 390 rpm on a turntable and observed under stroboscopic illumination. Mosquitoes were seen as reported by Kalmus and Hocking (1960) facing towards the center and walking towards it. Most of them collected near the center roughly 1 to 1.5 cm from it; fewer mosquitoes remained at the periphery. The centrifugal force at 1 cm from center was 1.7 g and 1.5 cm 2.5 g. As the dish continued to rotate more mosquitoes could be seen moving towards the center. For experiments with repellents the mosquitoes were taken in batches of 15, in a sucking tube, chilled for 1.5 minutes at 14°F and then their mesonota painted with repellent on a cold petri dish under a binocular microscope. All four repellents were tested. After treatment the mosquitoes were released in a cage and allowed to

recover. They were then introduced in the petri dish (50-60 of them) and made to rotate.

Under stroboscopic illumination it was observed that the mosquitoes did not collect in greater numbers near the center of the dish and the movement towards the center was less noticeable. The dish gave an appearance of a scattered distribution of mosquitoes as compared to a circular distribution near the center in the control. Quite a few (10-15%) faced directions other than the center.

TABLE 11 - Root mean square of angles of inclination of the body axes of A. aegypti to the vertical in the presence of repellents in a rotated petri dish in degrees. Means of five replicates ± standard errors.

| | Control | Repellents | | | |
		D.M.P.	D.E.T.	Rutger's 612	Indalone
Angle in degrees	0	43 ± 8.4	47 ± 13.3	50 ± 5.4	47 ± 13.3

In another experiment the mosquitoes themselves were not treated but a disc of 4 cm diameter (centrifugal force 3.5 g) was painted with repellent in the center of the dish. Mosquitoes (50-60) were introduced in the dish which was then rotated. It was observed that with an exception of one or two the mosquitoes remained outside the disc, sometimes facing towards it and sometimes turning away from it or walking around it. In yet another experiment when the diameter of the circle painted with repellent was increased to 6 cm (centrifugal force about 5 g) in 9 cm petri dish the same behavior was observed. Most of the mosquitoes remained outside the circle, although the non-treated peripheral belt around the repellent coated circle was only 1.5 cm wide.

Visual Responses

The optomotor and visual responses of mosquitoes have been studied by many workers. Kalmus (1958) reported that A. aegypti shows responses to the rotation of the plane of polarization of light. In a later study Kalmus and Hocking (1960, p. 19) observed swarming flight in A. aegypti close underneath a weak light source placed on top of a darkened cage, but the same was not observed when a much stronger light was made to pass through a red filter. Mosquitoes were also observed by these workers to aggregate near the margins of black objects when these were placed on top of a weakly illuminated cage.

The visual response of mosquitoes was also studied by Kennedy (1939) and Rao (1947). Kennedy reported that suspended mosquitoes orientated accurately towards a vertical black stripe on a white background. Presented with two stripes the mosquitoes faced one or the other stripe and not between the two. Rao (1947) reported similar findings with Anopheles maculipennis atroparvus van Thiel, and Culex (Culex) molestus Forskal

rendered flightless by the removal of the wings or by sticking them together.

To test the effect of repellents on the visual response of *Aedes aegypti* to black stripes, 20 female mosquitoes were taken in a glass bottle 12 cm tall and with a diameter of 3 cm. The inside of the bottle was lined with white nylon net to give the mosquitoes a good foothold. This bottle was placed inside a glass cylinder 14 cm high and with a diameter of 6 cm. The bottle and the cylinder were placed on a thick glass plate which was resting on a tripod stand. Under the glass was placed a 40 watt electric lamp which was covered all around with a cylinder of black paper so that light could go only upwards and light the bottle and the cylinder outside it uniformly from inside. In order that the inside of the cylinder be evenly illuminated, a filter paper was placed on the glass plate on which the outer cylinder and the inner bottle rested. The outer cylinder was divided into four quadrants and the alternate two quadrants were covered with black paper strips, each covering 90°. The remaining two quadrants were left uncovered, (figure 2).

As the outer cylinder was placed around the inner bottle containing mosquitoes and kept there for a short time, the mosquitoes inside moved and came to rest on the wall of the bottle facing the black stripes. The outer cylinder was then rotated 90° so that all the mosquitoes now faced uncovered portions of the cylinder. The mosquitoes moved again in the direction of the black stripes and again came to rest opposite to them. This behavior could be observed again and again. However, when the antennae were painted with any of the four repellents they showed complete indifference to the black stripes and did not move towards them as in the control.

The experimental data on the effects of repellents on **behaviour** are summarized in table 11A.

TABLE 11A - Summary of data on the effect of repellents on responses to stimuli.

Table / Page	Response	Effect
3/12 & 4/13	Sugar feeding	Inhibition
5/15	Mating	Partial Inhibition
6/18 & 7/19	Oviposition-site treated	Inhibition
	-tarsi treated	No Inhibition
8/20 & 9/21	To wind	Partial Inhibition (D.E.T. only)
10/23 & 11/24	Gravity	Inhibition
/ 24	Optomotor	Inhibition
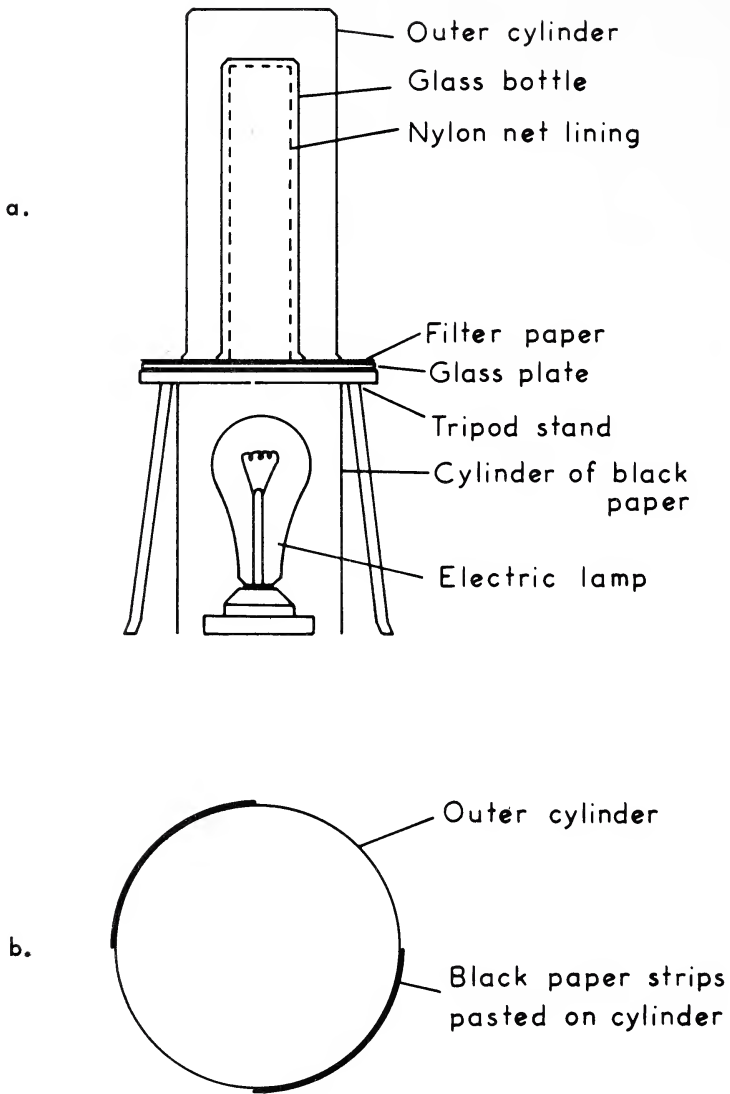
Figure 2. Diagrams showing: (a) a vertical section of the apparatus used for testing the visual response of A. aegypti females to black stripes in relation to repellents, and (b) a cross section of the outer cylinder.

EXPERIMENTAL - LIPOID SOLVENTS

Amongst the advocates of chemical theories referred to in a previous section, many have suggested lipoid solubility as a basis of olfaction (Cohn, 1924; Dyson, 1938; Dethier & Chadwick, 1947; Dethier, 1948). Experiments were conducted to examine the effect of fat solvents applied on the antennal chemoreceptors of *Aedes aegypti* females on their behavior towards a host.

Ten female *Aedes aegypti* eleven days old were taken for each experiment. The mosquitoes, which were fed on sugar solution only, were taken in a sucking tube and chilled for 1.5 minutes at $15^{\circ}F$. They were then placed on a filter paper on top of a cold petri dish and their antennae were washed with lipid solvents applied with a fine camel hair brush. The mosquitoes were then transferred to a clean petri dish lined with filter paper in a one cubic foot cage and allowed to recover. Thirty minutes after the operation a hand was introduced into the cage and the number of landings of mosquitoes on it was recorded for a period of 15 minutes. Mosquitoes were shaken off gently on landing and were not allowed to feed on blood. The antennae of controls were rubbed with a clean dry brush.

The observations are recorded in table 12, and show that the number of landings decreased very significantly on washing the antennal chemoreceptors with the lipid solvents. But whether the decrease in landings is due to the loss of lipids from the chemoreceptors, or due to the narcotic, anesthetic, or other effect of the solvents is uncertain.

TABLE 12 - Numbers of *Aedes aegypti* females landing on a hand in a 15 minute period after treatment of the antennae with lipoid solvents. Means of three replicates ± standard errors.

Control	Acetone	Ether
174 ±8	52 ± 12	43 ± 13

DISCUSSION

The action of repellent chemicals on mosquitoes has no specificity for blood feeding behavior. It has been shown that repellents in the vapor phase have the following effects on *Aedes aegypti*. They inhibit feeding on both blood and sugars, reduce the mating rate, and cause rejection of oviposition sites. The repellents also affected orientation to gravity and centrifugal force and the visual response to black stripes.

Mosquitoes became quiescent and less active when repellents were applied on them. This slowing down of motor activity suggests the external stimuli normally acting on the mosquito are perhaps blocked or interfered with by the repellent. As there is no delay in the effect of repellents on the behavior of mosquitoes, that is, protection is obtained immediately these materials are applied, their action on the insect may be assumed to occur at the surface of the body. Repellents have not been shown to penetrate rapidly into the body where they could act on

the nerve synapses or the central nervous system, nor have they been shown to affect the muscular system directly. It thus seems unlikely that they act by blocking the nerve impulses or the motor response. The most probable action seems therefore to be the blocking of reception of stimuli at the receptor site.

Somewhat different behavior in relation to repellents of another kind has been described by Kennedy (1947). He studied the effects of contact with DDT on the activity and distribution of mosquitoes. He argued from his experiments that a variety of reactions may give rise to repulsion. Reactions may occur at a distance or only after contact with a repellent surface. The contact stimuli may be mechanical or chemical. The reactions may take the form of an increase of merely random activity or they may be directed away from the surface. They may be quick or slow to appear and weak or strong in expression. In contrast to my findings of reduced activity in his work an increase in activity was found.

The factors that attract mosquitoes to the host have been reviewed above. The mode of action of insect repellents can be best understood when studied in relation to these factors.

The effects of repellents on the evolution of carbon dioxide and moisture from a human arm, and the correlation of this evolution with the natural attractiveness of human beings and protection time of repellents were studied by Gouck and Bowman (1959) at Orlando, Florida. In their experiments, repellents applied to the arms of three subjects reduced the CO_2 emitted by 9 to 14 per cent but they concluded: "Although these reductions are considerably greater than the differences between untreated arms (4%) they are not great enough to indicate that the mode of action of these repellents is based upon the retardation of carbon dioxide evolution". The repellents used were, dimethyl phthalate, diethyl toluamide and ethyl hexanediol. With regard to the moisture collected from untreated and repellent treated arms they concluded: "The quantities from the arms of all subjects varied from day to day but in most individual tests the two arms agreed within about 5 per cent indicating that no real difference in the amount of moisture evolved was caused by application of repellents." They believed that the protection time is governed by the rate of loss of repellent from the skin by absorption and evaporation. Peters and Kemper (1958) have shown that there are no considerable temperature differences between repellent treated and untreated parts of the skin.

In the light of these findings it can be said that repellents affect the reception of these stimuli rather than the stimuli themselves. This supports the hypothesis advanced that repellents affect many kinds of behavior of mosquitoes by interfering in the reception of many different kinds of stimuli.

Search for chemical factors other than carbon dioxide attracting mosquitoes to the host has claimed the attention of many workers. The findings of Shaerffenberg and Kupka (1951) and Burgess and Brown (1957) have indicated that attractive factors other than carbon dioxide are present in the vapor from mammalian blood and body exudations. A distillate obtained from mammalian blood by Shaerffenberg and Kupka

(1959) proved highly attractive to *Culex pipiens* L. Rudolfs (1922) found benzoic acid, dilute ammonia, phenylalanine, alanine, aspartic acid, cystine, and hemoglobin to be attractive to *Aedes sollicitans* Walker and *Aedes cantator* Coquillett, but Reuter (1936) found the last six materials unattractive to *Anopheles maculipennis atroparvus*. Brown and Carmichael (1961) reported that L-lysine and L-alanine were attractive to *Aedes aegypti*. The effect of repellents in association with these chemicals found to be attractive remains to be studied.

Travis and Smith (1951) evaluated dimethyl phthalate, indalone, and ethyl hexanediol against *Aedes aegypti* besides other mosquitoes, and found average repellent times (i.e., times in minutes from application of the repellent to the first bite) as follows: ethyl hexanediol - 331 minutes, dimethyl phthalate - 247 minutes, and indalone - 111 minutes. Although the results of my experiments are not strictly comparable with those of Travis and Smith (1951) for I worked with a different culture of mosquitoes and at a different time and place, the mosquitoes fed on blood much sooner after treatment when repellents were applied on the mosquito receptor sites. For example, about 33 per cent of mosquitoes fed on blood within 10 minutes after application of dimethyl phthalate on both antennae. When diethyl toluamide, indalone, and ethyl hexanediol were separately applied on both antennae, some of the first bites were recorded after 10 minutes. The reason for this behavior is perhaps the more rapid adaptation of the receptors to the repellents because of the greater concentration gradient resulting from their application on the receptors themselves. In this way the threshold for reception of repellents increased greatly but that for other stimuli remained the same. The sequence of stimuli and responses leading to blood feeding therefore remained unaffected. But this is, of course, incompatible with the hypothesis that repellents block all receptors.

The presence of separate chemoreceptor neurons mediating acceptance and rejection is assumed from the study of labellar chemoreceptor cells of *Phormia regina* . These cells have been the subject of coordinated behavioral, histological, and physiological study. A chemosensory hair of the labellum of this blowfly was described by Dethier (1955) as a hollow extension of the body cuticle possessing two distinct lumina. The chemosensory hair has been shown to be associated with three bipolar neurons, two of which send distal fibers to the terminal papilla by way of the thick-walled lumen of the hair. Dethier (1955) concluded that one of these neurons mediates acceptance while the other mediates rejection. On electrophysiological studies one of the two neurons was later designated the L fiber (for large spikes which responded to salts and the other the S fiber (for small spikes) which responded to sugars (Hodgson et al. 1955; Hodgson and Roeder, 1956). Wolbarsht and Dethier (1958) were able to detect the spikes of the third neuron which terminated in a process at the base of the hair. It was designated M for mechanoreceptor. Evans and Mellon (1962) have now detected spikes from a fourth neuron which responds to water.

In the course of electrophysiological studies of chemoreceptor hairs it has been shown that when mixed stimuli are applied there is an interaction between activity in the L and S fibers (Hodgson, 1956 ,

1957; Morita, 1959; Sturckow, 1959). Hodgson (1957) found that the presence of S impulses is accompanied by a decrease in L impulses and conversely the S spikes decrease when the L fiber is stimulated. My experiments show that the repellents block the reception of attractant and other stimuli. This assertion needs to be confirmed by electro-physiological methods.

Mosquitoes with antennae painted with diethyl toluamide landed, walked around, and even probed on an arm also treated with the same repellent but did not feed on blood. This may be explained in one of two ways. It may be that the piercing of the skin by the mosquito is induced by some chemical factor on the skin which was neutralized by the application of the repellent or, it may be due to the effect of repellent on the action of thermoreceptors or contact chemorecptors which induce feeding on blood. The latter explanation would be more in conformity with the findings that repellents interfere with the reception of all kinds of stimuli affecting the total behavior of mosquitoes.

The study on blood feeding when repellents were applied on parts of the mosquito revealed that of the four repellents dimethyl phthalate has the greatest effect on blood feeding behavior when it is painted on the tarsal receptors and the smallest effect when it is painted on the receptors of the antennae. As is known, the olfactory receptors are located on the antennae and the contact chemoreceptors mostly on the tarsi of the mosquito. Dimethyl phthalate, which has the highest boiling point and hence the lowest vapor pressure, may, for this reason, have more effect than the other repellents through the tarsal chemoreceptors in the liquid phase but less than these through the olfactory receptors of the antennae where it has to act in the vapour phase which is at a lower concentration.

That repellents also acted as irritants was evident from the intense wriggling activity of the mosquito when repellents were applied on the proboscis and from the vigorous cleaning of repellent from the antennae with the tarsi of the fore legs. This evident awareness of the presence of an irritant chemical indicates the existence of receptors sensitive to it, perhaps those of the common chemical sense. It may well be that these are the only receptors not blocked by repellents.

In the vapor phase repellents were found to inhibit landing of mosquitoes. This was observed in experiments on blood feeding, sugar feeding, oviposition, and air flow. In the liquid phase, however, the repellents showed more irritant and some toxic effects, and the mosquitoes showed considerable decrease in locomotor activity, in part on account of preoccupation with attempts at cleaning off the repellents.

Repellents have been defined as compounds which elicit an avoiding reaction (Dethier, 1956b). While the four materials studied may all do this, this is by no means their only effect and may not, indeed, be the most important one.

ACKNOWLEDGEMENTS

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Book Review

LINDROTH, C.H. 1963. The ground-beeltes of Canada and Alaska.
Part 3. Opuscula Entomologica, Supplementum XXIV, pp. 201-408,
Figs. 102-207. Zoological Institute, University of Lund, Lund, Sweden.
Price - 35 Swedish crowns.

This portion of this work, the second to be published, includes
the last part of the taxonomic treatment of the genus *Trechus* , and a rev-
ision of the bembidiine genera *Asaphidion* Gozis (three species), *Bembidion*
Latreille, and the monotypic genus, *Phrypeus* Casey. The treatment of
Bembidion occupies 200 of the 207 pages. This volume is based on an
examination of the relevant material stored in the major European and
North American museums and private collections, **and on the extensive**
collections of Lindroth.

As in part 2, Lindroth provides for each species a succinct
synonymy, a synoptic description, and data on type locality, ecology, and
geographical distribution.

The text is straight-forward, simple English. The resulting
clarity of expression illustrates very well the author's thorough knowl-
edge of his subject.

The illustrations are excellent, and those of the entire insects
are among the best ever executed of carabid beetles. For many of the
species, the internal sac of the male genitalia, with its complex folds
and peculiarly shaped sclerites is illustrated, in the infolded position.
Also provided are simple, clear-cut line drawings of various other
structures. All drawings were made by the author himself.

The treatment of the genus *Bembidion* is the dominant feature
of this volume. The 193 species, 31 of which occur in the United States
only (excluding Alaska), are arrayed in 48 groups. An additional six
extra-limital species are included in the key to species, but are not
treated elsewhere in the text. For each group, a brief diagnosis is
given, as well as the subgeneric name that would apply if the author chose
to use the category subgenus. Twenty-five new taxa are described, of
which four are ranked as subspecies. Of the new species, the type
localities of six are in the United States (excluding Alaska). Although
the work deals primarily with the Canadian and Alaskan fauna, Lind-
roth treated all of the known North American species for a number of
the species groups.

Bembidion has long been regarded as the most difficult and complex
genus of carabids in North America, and the justification for this opinion
is perhaps best illustrated by the large number of synonyms listed -
165 - of which 159 were proposed by one author, Colonel Thomas Lincoln
Casey. (By way of contrast, 21 Casey species are recognized as valid,
and his names are also used for another two species, as a result of the
first-used names being junior homonyms). The synonymy is based upon
study of the type specimens by Lindroth, and the facts should settle any
doubt about the value and quality of Casey's work in the Carabidae.
Hayward's revision of 1897 (Trans. Amer. ent. Soc. , vol. 24) was also
grossly inadequate. Lindroth's extensive knowledge of the European

species of *Bembidion* , plus his thorough familiarity with Netolitzky's fine study are factors which contributed in an important way to the success of the study of the North American species. Thanks to this revision, it is now a relatively simple task to determine any specimen from Canada or Alaska.

The two keys for identification (one to species groups, and one to the species) are easy to use. This statement is based on personal experience gained by identifying several thousand specimens, representing a substantial portion of the species. Each couplet in the keys consists of a pair of clear-cut alternatives, and there are no complicated "either-or" statements. One of the features facilitating use of the long key to species (225 couplets) is that the numbers of those couplets which set off a large number of species are in bold face. In spite of these good features I have three criticisms to make regarding the keys: a. no attempt was made to relate directly the species-group key to the species key; b. names of authors of species were not given in the key; c. page references to the text were not given for the Canadian and Alaskan species. However, these are minor points, and the last one is largely taken care of by the number which is assigned to each species in both key and text.

In a key of this length, it is almost impossible to avoid errors, and it is with regret that the following omissions of species are noted: 64. *nigrum* Say; the species of the *incrermatum* group-103 *incrermatum* LeConte, 104. *immaturum* Lindroth, and 105. *graciliforme* Hayward; and *humboldtiense* Blaisdell, p. 305.

The fact that only a few subspecies were described or recognized may suggest that the author is unaware of current taxonomic theory. Such, however, is not the case. Lindroth notes carefully geographical variation where he finds it, but he describes as subspecies only those populations which are clearly geographically isolated from their closest relatives, and which differ markedly from them. He avoids naming populations which are segments of clines, and thus avoids proposing a lot of trinominals which will subsequently have to be synonymized.

A search through the work for indications of modern techniques of analysis will prove fruitless. One does not find complex graphs, charts, or long tables, and only very few simple statistical parameters are indicated. However, the study does not suffer from this seeming lack. This seems to me to show that a major attribute of a good taxonomist is the ability to interpret correctly carefully chosen, accurate observations. This is not to say that the study of the genus cannot be pursued profitably with more sophisticated techniques, but rather that I doubt that such techniques would have provided, at the present level of understanding, much more than Lindroth was able to state using the methods of analysis that were in use in the time of Linnaeus. This illustrates that the difference is unimportant between 'modern' as opposed to 'old fashioned' taxonomy; the distinction should rather be made between 'good' and 'poor' taxonomy.

Regarding classification of *Bembidion*, I think the author is mistaken in using only a single infra-generic category, namely 'group'. In a genus of this size, several infra-generic categories are required to point out the similarities and differences among the species: subgenus, species

group and sub-group, at least, However, Lindroth states that such a classification should be proposed on the basis of a study of the world fauna, and perhaps he is right.

The work has, so to speak, opened the door to the study of North American *Bembidion*. It provides a basic classification, which can be easily modified, as required. It shows clearly how diverse the genus is. The task of completing the revision of the North American species will be a pleasure. Because of the marked ecological specialization of many of the species, the genus should provide valuable material for the study of the origins of adaptations. Also, the numerous species and their wide distribution in North America, should provide fertile ground for the development of zoogeographic studies. And, returning to description of structures, one should remember that the immature forms are virtually unknown. Lindroth has provided an excellent platform from which to launch further studies, and it is to be hoped that such studies will be made in the near future.

Carl Lindroth brought to this work a feeling for these fascinating little creatures which is best described as deep affection. And this, combined with unrivalled knowledge, superb talent, and hard work on the part of the author, has provided us with the finest taxonomic treatment of a group of carabid beetles ever produced.

George E. Ball

A

Ent

Quaestiones

entomologicae

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CONTENTS

Editorial — Beastly teachers

Teachers, they say, are a necessary evil; beastly people, teachers; pedantic, dogmatic, intolerant. If this is the nature of the beast, should we not take Wordsworth's advice and 'let nature be our teacher'? There could be no better field than entomology in which to put this into practice; at least we should run no risk of a shortage of teachers.

It is difficult to arrive at a reasonable estimate of the world population of entomologists, because they are difficult people to define and still more difficult to regiment (praises be!). If one supposed that for every one attending an International Congress, ten stay at home - or more likely go out collecting - there must be around 20,000. If Canada has as many per head of population as any country, as has been claimed, the figure may be 50,000. Let us average these two figures; if we have 35,000 entomologists, this would allow 22 described species of insect per entomologist, or if we accept C. B. Williams' estimate of the world population of insects at 10^{18}, about 3×10^{13} insects per entomologist; a rather unusual staff/student ratio.

Insects are certainly pedantic, dogmatic, and intolerant, and should therefore make good teachers. And as teachers of entomology they must surely be immune to the fashionable accusation directed at school teachers - that they are good teachers but have nothing to teach, if not to the reciprocal retort often aimed at university teachers. Perhaps this is the proper role of human teachers of entomology - to help the insect teach the student, or to help the student to learn from the insect. Certainly if one had to choose between insects, books, and entomologists, from which to learn, the choice would be in the order given. Perhaps more than any other science, biology in general and entomology in particular must be taught from the organisms they are concerned with, in the field and in the laboratory. Many of us get into the bad habit of reaching for a text when in doubt about some point of insect structure, when we could just as

easily reach for an insect - a much less fallible adviser. The habitual reference of questions back to the insect might even help us in our difficulties in keeping up with the literature; it would certainly give us a surer foundation of knowledge from which to judge whether, in any particular paper, we need to read on. In addition to the rather negative qualities we started out with, insects are ubiquitous, lively, versatile, unobtrusive, fertile, and unequivocal. There is little more one could ask of a teacher.

One of the interesting advantages of an insect teacher of entomology as compared with a human teacher, is that he can fulfil many of his functions even after death, especially if well preserved. Indeed it is in large part the readiness with which they may be acquired in the first place and preserved in the last place, that makes insects so much more valuable than many other groups of organisms in the teaching of other branches of biology. Their only limitation lies in their inability to teach the structural detail of other groups - unfashionable stuff these days anyhow.

There is a tradition of great teachers of entomology extending back to the early years of the science itself. Surely a place in this roster has been earned at least by two species of cockroach, by a fruit fly, and by mealworms and flour beetles.

Brian Hocking

THE FUNCTIONAL MORPHOLOGY OF THE MOUTHPARTS OF SOME MOSQUITO LARVAE

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Homologies of the parts of the maxilla and the labium of mosquito larvae were studied. The name cardobasistipes is proposed for the triangular sclerite latero-posterior of the maxilla, previously known as the cardo or the palpifer. The numbers of serrations on the prementum and submentum were found to be of taxonomic value. The sequence of mouthpart movements of filter feeding and browsing species, and the progress of food particles from the feeding current into the mouth were observed. Differences in stiffness were found among the setae in different positions on the mouthparts. These differences were confirmed by staining the cuticle with Mallory's triple stain and are correlated with the functions of the setae during feeding. Flexible serrations at the tips of the labral brush hairs are used for raking food particles in most of the browsing species of Aedes *and* Culiseta *studied. When in pond water neither the browsing nor the filter feeding larvae select the type of food they ingest. Feeding behaviour of the predatory larvae of* Chaoborus americanus *(Johannsen) and* Mochlonyx velutinus *(Ruthe) was observed.*

INTRODUCTION

The mouthparts of a mosquito larva occupy a large portion of its head; their structure is degenerate. In this work emphasis is placed on the homologies of the parts of the maxilla and the labium, on the structure and function of the labral brushes and on the type and size of food particles ingested by the larvae.

The problems of homologies of the mouthparts did not occupy the early biologists who lacked adequate equipment for detailed study of minute structures. Hooke (1665) drew a mosquito larva, but he did not interpret all the parts of its anatomy accurately; for example, he labelled the external opening of the respiratory siphon as the anus. He further said about the "Water - Insect or Gnat": --"It is suppos'd by some, to deduce its first origin from the putrifaction of Rain Water. . ." He wrote that the larvae can move gently through the water by moving their mouthparts, and "eat" their way up through the water.

Reaumur (1738) described and illustrated the external features of a mosquito larva which seems to be a *Culex* species (*pipiens* according to Shannon, 1931). He gave an accurate description of the function of the labral brushes and described browsing and filter feeding activities of larvae.

The best known studies on mosquito larvae in the 19th century

are those of Meinert (1886) and Raschke (1887) who discussed larval morphology, function of mouthparts, and some of the habits of larvae and adults.

The names used by authors for the mouthparts of mosquito larvae are summarized in table 1. The following authors also referred to some mouthparts by specific names: Miall (1895), Johannsen (1903), Mitchell (1906), Puri (1925), Montchadsky (1945), and Cook (1956). A more complete list of literature on this subject is included in my thesis (Pucat 1962). It is evident that there is disagreement on the homology and nomenclature of certain mouthparts. There is less disagreement on the function of these parts, but this has not been studied exhaustively.

Classification of Feeding Habits

The structure of mouthparts, the method of feeding, and the habitat of the larvae are inter-related. On the basis of these factors culicine larvae have been classified into filter feeders, browsers, and predators (Surtees 1959).

It has been found convenient to follow this classification since it is based on morphological and functional characteristics. The criteria may be summarized as follows:

Filter Feeders – are larvae which strain out food particles from the water, such particles being sufficiently small to pass directly into the digestive tract without undergoing any further breakdown. Their salient morphological characters are: long, fine, unserrated labral brushes, large maxillae bearing many fine setae, small weakly chitinized mandibles, a weakly chitinized submentum possessing a large number of very small teeth and, associated with these features, large sub-apical tufts of setae on the antennae (Surtees 1959). These structural features were recognized by Wesenberg-Lund (1920) in several Danish species of mosquitoes. Nuttall and Shipley (1901) described in detail the function of the labral brushes of a filter feeder, an unnamed *Anopheles* species.

Feeding action similar to that observed by Nuttall and Shipley was also observed by Bekker (1938a, b) in *Anopheles maculipennis* Meigen, and by Renn (1941) in *Anopheles quadrimaculatus* Say and *Anopheles crucians* Wiedemann. Renn referred to the characteristic anopheline feeding method in which the floating particles are drawn straight towards the mouth as "interfacial" feeding. However, sometimes anopheline larvae employ a feeding method common to the larvae of other genera of mosquitoes in which the particles move in converging curved lines, and this Renn calls "eddy" feeding.

Browsers – abrade solid material, the particles of which require further manipulation by the mouthparts before entering the digestive tract (Surtees 1959). Mouthparts of this type have been described by Mitchell (1906), Howard, Dyar, and Knab (1912), Wesenberg-Lund (1920), Surtees (1959), Snodgrass (1959), Christophers (1960), and Clements (1963). All authors agree that browsing larvae are usually bottom feeders.

The labral brushes as well as the maxillary and mandibular bristles are shorter and stiffer than in the filter feeders. As Mitchell (1906)

pointed out, in brushing over debris at the bottom of a pool very long, slender hairs would be a disadvantage. Mandibles are used to manipulate any large particles that come into the feeding stream, and the submentum is used as a secondary grasping organ. The swimming position is usually at an angle of about 45° to the substratum. Morphological gradations occur between typical filter feeders and browsers (Wesenberg-Lund 1920, Surtees 1959).

Predators - have the labral brushes strongly chitinized. The role of the maxillae has been suppressed and the mandibles are the principal mouthparts. These are very large with strongly chitinized claws and take up most of the oral region of the head capsule. Associated with the strong claws are large, stiff spines which also aid in grasping the prey. This is true of the larvae of *Chaoborus* and *Mochlonyx* (Schremmer 1950, Peterson 1951, Cook 1956, and others). The submentum in all predatory species is well developed, the teeth being large and generally pointed. The increase in the strength of the submentum is associated with a reduction in the number of teeth and mouth brushes. Predatory larvae have large prehensile antennae which aid in grasping prey.

Evolution

Montchadsky (1937) has considered the environmental adaptation of larval and adult structures and behavioral characteristics important in classification. The type of feeding is a factor correlating the processes of evolution of larval and adult mosquitoes.

The Anophelinae and Culicinae have mostly plant-feeding larvae and blood-sucking adults (Montchadsky 1937, Hennig 1950). However, the Toxorhynchitinae and the culicine subgenus *Lutzia* have reversed their type of feeding; the larvae lead a predatory life, but have structures which indicate a previous adaptation to a vegetarian type of feeding. The adults of these mosquitoes either feed on plant juices (but carry traces of previous ability to suck blood), or appear to be optional blood feeders (Montchadsky 1937). In the Chaoboridae the adults are plant feeding while the larvae are predatory. Two lines of adaptation to predation are known: the surface film feeders such as *Eucorethra*, and the pelagic feeders such as *Chaoborus*.

In the initial stages of evolution of the mosquitoes either there was a change in the type of feeding of the adults (transition to blood feeding in the subfamily Culicinae), or of the larvae (the transition to predation in the Chaoboridae). According to Montchadsky (1937) these changes were provoked by certain changes in the nutritional requirements for the ripening of the sexual organs. If adequate food containing high quality protein is eaten by the predatory larvae, it is not then required to be eaten by the adults which may be vegetarian. On the other hand, non-predatory mosquito larvae do not obtain adequate high quality protein, so that the adults of these species must have it from the blood of vertebrates.

TABLE 1 - Summary of names which have been used for some mouthparts of mosquito larvae.

Author	Labral Area						Maxilla		Labium		
Meinert 1886 *C. annulatus*	clypeus	whirling organ	scutum of 1st metamere				internal lobe	external lobe		underlip	
Raschke 1887 *C. nemorosus*	upper lip	Strudel-apparat					maxilla			underlip	
Giles 1902 *A. rossi C. fatigans Mochlonyx Chaoborus*	labrum	whorl organ					maxilla			lower lip	
Theobald 1901 *Anopheles Megarhinus*		whorl organ brush					maxilla			labial plate	
Nuttall & Shipley 1901 *Anopheles*	clypeus	brush					maxilla			lower lip	
Thompson 1905 *Culex*		flabella	palatum	flabellar inner retraction insertion	flabellar outer retraction insertion	epipharynx	maxilla		hypopharynx labium	mental sclerite	
Imms 1907,1908 *Anopheles*	clypeus	brush		chitinous apodeme of epipharynx		epipharynx	maxilla		hypopharynx	labial plate	
Wesché 1910 *Culex*	brush						maxilla		labium (pre-mentum)	lower lip	
Howard 1912 *Culex Aedes Anopheles*	labrum	mouth brush flabella	palatum			epipharynx	maxilla		labium hypopharynx	mental sclerite	hair fringed plate
Wesenberg-Lund 1920 *Culex Culiseta Aedes*	labrum	flabella	palatum	apodeme			maxilla		labium hypopharynx	mental sclerite	hair fringed plate
Salem 1931 *A. fasciatus*	labrum	feeding brush	palatum	apodeme	clypeo-labral suture	epipharynx	maxilla	first segment of palp	labium	mentum	submentum
Bekker 1938	clypeus	flabella	antero longitudinal	transverse		epipharynx	maxilla		labium		

Author / species												
Cook 1944, 1949 *Culiseta incidens*	labrum	labral brush	palatum	messor	messorial apodeme		palatal bar	stipes	palpifer	pre-mentum	sub-mentum	aulaeum
Matheson 1944 *Anopheles*	labrum	mouth brush	palatum					maxilla		pre-mentum	mentum	mentum
Farnsworth 1947 *A. quadrimaculatus*	labrum	labral brush	median labral brush	messor	messorial apodeme		palatal bar	maxilla		hypopharyngeal body	inner tooth of mentum	outer tooth of mentum
Schremmer 1949 *A. maculipennis*	labrum						epipharynx Apparat	maxilla			labium	
Chaudonneret 1951 *C. pipiens*	labrum			messor			epipharynx	maxilla				
Foote 1953 *C. percator C. atratus*	labrum	mouth brush	mouth brush	messor			palatal bar	maxilla		pre-mentum	mentum	aulaeum
Shalaby 1957 *Aedes Culex Culiseta*	pre-clypeus	lateral labral brush	palatum	labral brush apodeme	posterior apodeme	transverse bar	epipharyngeal sclerite	lacinia galea cardo - stipes	palpifer	hypo-pharynx	para-glossa	glossa
Menees 1958 *A. quadrimaculatus*	labrum	mouth brush	median brush	torma		anterior intertormal bar	posterior intertormal bar	lacinia galea stipes	cardo	labio-hypopharyngeal body	inner tooth of mentum	outer tooth of mentum
Snodgrass 1959 *Aedes Culex Culiseta Anopheles*	labrum	lateral feeding brush	median brush	torma	apodeme	epipharyngeal bar	epipharynx	stipes	cardo	labium hypopharynx	hypo-stomium	aulaeum
Surtees 1959 *Culex Aedes*		mouth brush	mouth brush					maxilla		mentum		
Christophers 1960 *A. aegypti*	pre-clypeus	flabellum	palatum	apodeme	stirrup apodeme	posterior palatal bar	epipharynx	distal part of maxilla	palpifer	labium hypopharynx	mental sclerite	
Jones 1960 *A. quadrimaculatus*	labrum	lateral labral brush		messor			palatal bar	maxilla		pre-mentum	mentum	sub-mentum
Present work	labrum	lateral labral brush	median labral brush	torma	posterior tormal apodeme	transverse bar	epipharyngeal bar	lacinia disti-stipes	cardo basistipes	pre-mentum	mentum	sub-mentum

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Salem 1931 *A. fasciatus*	labrum	feeding brush	palatum	apodeme		clypeo-labral suture	epipharynx	maxilla	first segment of palp	labium	mentum	sub-mentum
Dekker 1938	clypeus	flabella	antero median apodeme	longitudinal lower		transverse	epipharynx	maxilla		labium	mentum	sub-mentum
Marshall 1938 *Aedes Culex Anopheles*	pre-clypeus	mouth brush					epipharynx	maxilla		hypo-pharynx	mentum	sub-mentum
Cook 1944, 1949 *Culiseta incidens*	labrum	labral brush	palatum	messor	messorial apodeme		palatal bar	stipes	palpifer	pre-mentum	sub-mentum	aulaeum
Matheson 1944 *Anopheles*	labrum	mouth brush	palatum					maxilla		pre-mentum	mentum	
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MORPHOLOGY OF THE HEAD AND MOUTHPARTS OF MOSQUITO

LARVAE

The mouthparts of mosquito larvae were compared with the mouth-
parts of larvae of other Nematocera, Mecoptera, and other panorpoid
groups, or with published descriptions of them.

Procedures

Two species of mosquito, *Aedes aegypti* (L.) and *Culiseta   inornata*
(Williston) were reared in the laboratory, so that fresh specimens of
these species were almost always available.   Rearing methods of
Trembley (1955) and McLintock (1952) were followed. Specimens from
the field were also observed alive and dissected in the laboratory. Since
larvae were available in abundance, dissected heads were mostly studied.
The dissections were done in glycerine. Hoyer's mounting medium and
neutral Canada Balsam were used for mounting the mouthparts. Eosin-
water solution was used for staining dissected muscles, and modified
(Peterson 1960) Mallory's triple stain for larval head cuticle.   The
mouthparts were boiled for 15 minutes in an 8% aqueous solution of
KOH before staining.

Manton (1958) commented on the staining reaction of cuticle
with Mallory's.   She concluded that sclerotized non-staining exocuticle
is unstretchable when thick, that orange and red-staining cuticle are
progressively less fully slcerotized, less rigid, and more elastic than
the non-staining cuticle, and that blue-staining cuticle is fully flexible,
more stretchable, but less elastic.

The structure of the heads of the larvae of *Aedes fitchii* (Felt and
Young) and *Culiseta inornata* was studied in detail, and other species (table 2)
were compared with them. Larvae of a *Chironomus* species, and of *Mochlonyx
velutinus* (Ruthe) and *Chaoborus americanus* (Johannsen) were also examined.

The Head Capsule

The largest sclerite in the head capsule of a mosquito larva is the
frontoclypeus, which extends over most of the head surface dorsally.
The genae are lateral, the postgenae postero-lateral; they extend vent-
rally to complete the head capsule (figs 1, 2). The median ventral part
of the united postgenae, posterior to the mouth, has been given various
names. I consider it as the subgena. It is bounded by two lines of
cuticular thickening ridges which are known variously as the submental-
postgenal sutures (Shalaby 1956 and 1957a, b, c, d) hypostomal sutures
(Menees 1958a, Christophers 1960), and thickening ridges (Snodgrass
1959). I agree with Snodgrass' interpretation of the homologies of the
ventral head sclerites. In homologizing these sclerites of the mosquito
larva Snodgrass digresses to discuss the ventral head sclerites of other
insects, especially insects in which a trend toward a ventral elongation
of the postgenae is evident. As examples he cites certain beetles in which
the entire labium with a gular addition to the submentum is enclosed
between the postgenae.   He states, however, that this condition is not
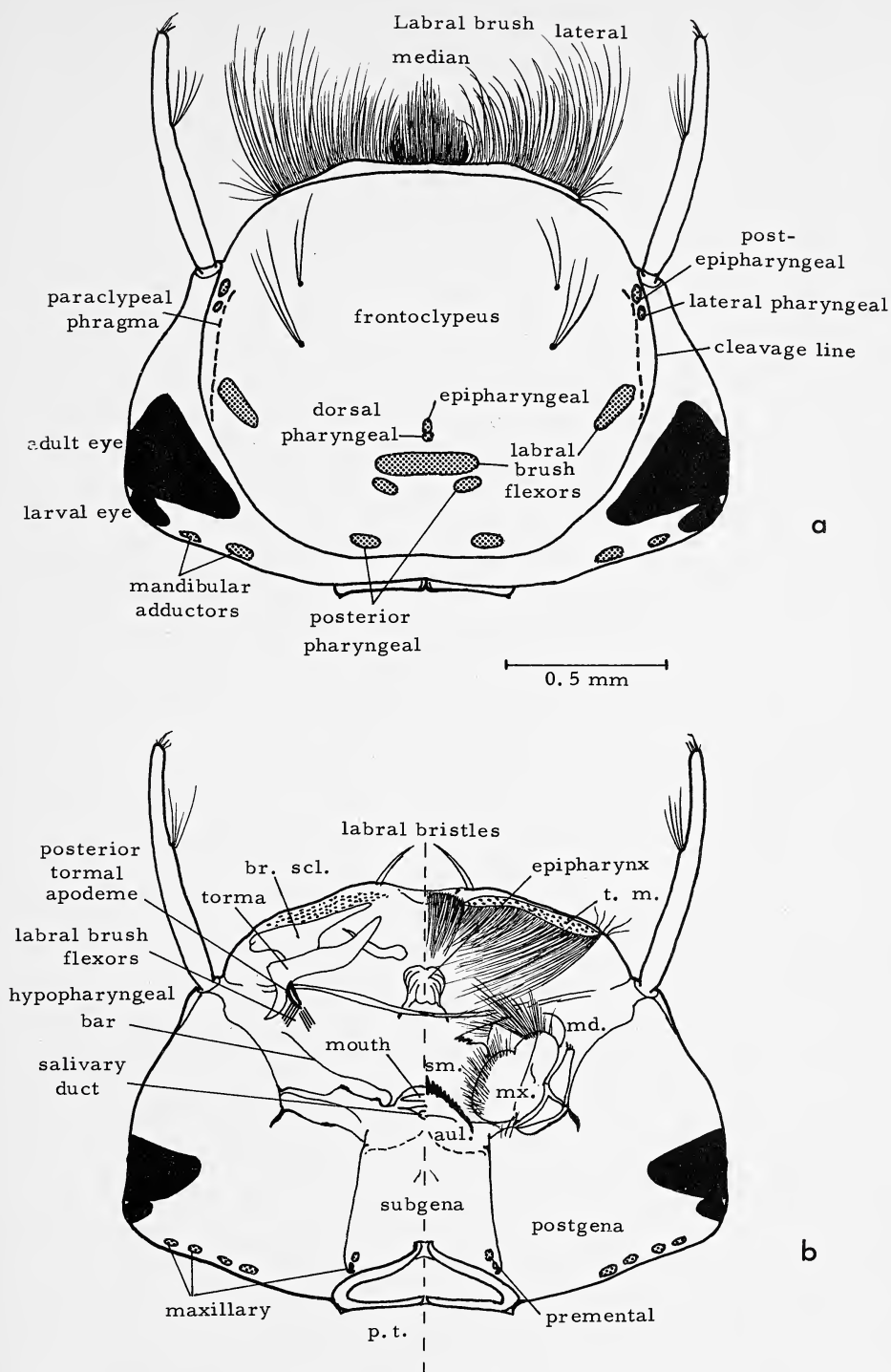
Fig. 1. The head of *Aedes fitchii* (F. & Y.) larva, (a) dorsal view showing muscle origins and extended labral brushes, (b) ventral view with brushes retracted and mouthparts removed from right hand side. mx. maxillae, md. mandible, sm. submentum, t.m. tessellated membrane, aul. aulaeum, p.t. posterior tentorial pit. Muscle attachments stippled.
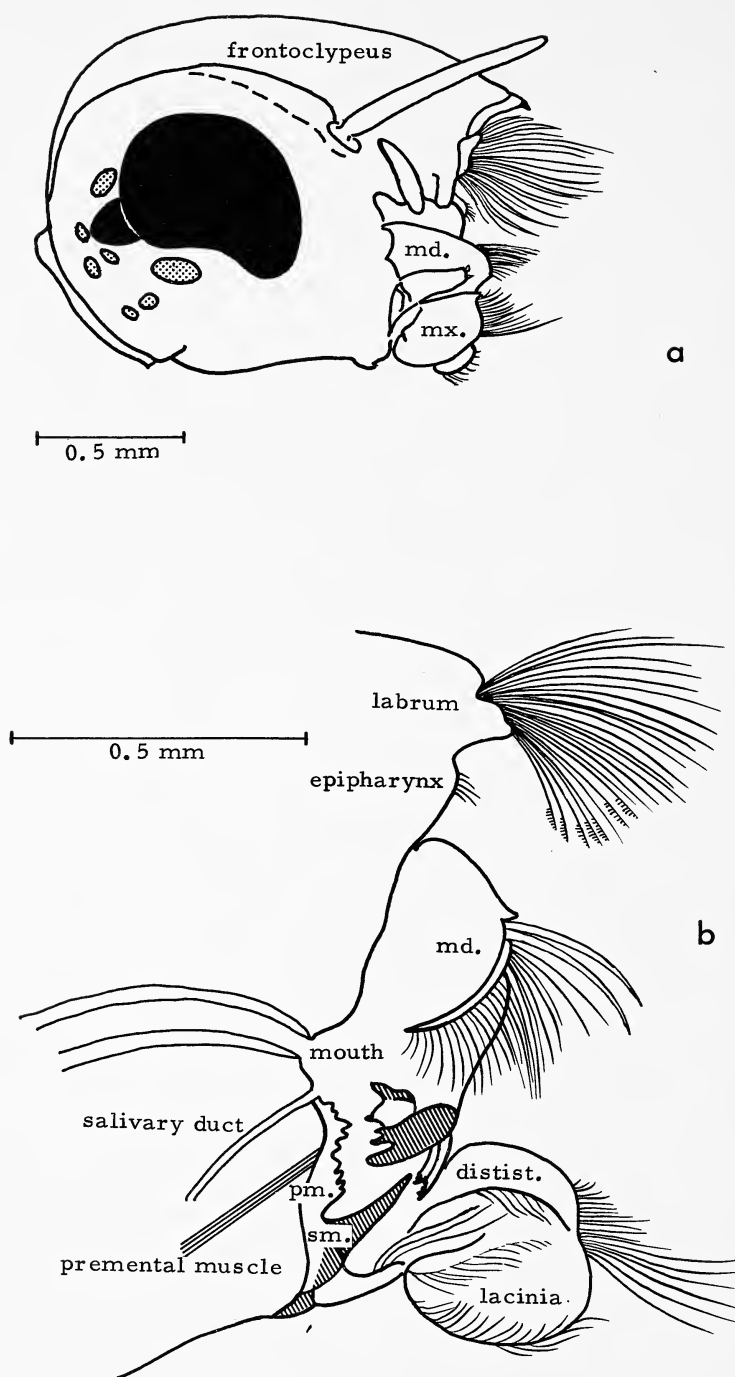
Fig. 2. (a) Lateral view of the left side of the head of *Aedes fitchii* (F. & Y.) larva. (b) Sagittal section through the mouthparts of *Aedes fitchii* larva. md. mandible, mx. maxillae, pm. prementum, sm. submentum, aul. aulaeum, distist. dististipes. Muscle attachments stippled.

represented in mosquito larvae. More commonly, the postgenae come to-
gether medially and displace the labium. A final stage in the displacement
of the labium is seen in the larvae of Chironomidae where the labium has
become greatly reduced and is hidden from below by a median hypostomal
lobe of the united postgenae.

A similar process of closure and elongation of the postgenae and
reduction of the labium occurs in nematocerous larvae as discussed by
Anthon (1943), Hennig (1948, 1950, 1952), and Snodgrass (1959). In
the larvae of the primitive rhyphid *Olbiogaster* the small postgenal lobes
are posterior to the submentum of the labium (Anthon 1943). In tipulid
larvae, described by Vimmer (1906) and other authors, as well as in
other nematocerous larvae the genae are completely united ventrally
and the labium is dorsal to the subgenal lobe. In the mosquito larva,
to distinguish the central area between the thickening ridges of the genae
Snodgrass (1959) named it the subgena, and the areas laterad of the
ridges the postgenae. I use this nomenclature.

Cook (1944a, b, 1949), following Ferris's (1947) and Henry's
(1947) theories of the segmentation of the arthropod head, considered the
postgenae and the subgena as parts of the maxillary segment. Shalaby
(1957) considered the apical part of the subgena as the mentum and the
remainder as the submentum. As evidence for this idea Shalaby
referred to Wheeler's (1893) embryological work in which the latter
observed that the rudiments of the second pair of maxillae on the sides
of the embryonic body give rise to the labium in the embryos of the locust
Xiphidium ensiferum Scudder, in *Gryllus luctuosus* Serville, and in *Stagmomantis
carolina* (Johannsen). Shalaby believed that the median suture present
on the ventral sclerite of the head of *Culex molestus* Forsk. larva is due
to incomplete fusion of the embryonic rudiments of the second maxillae.
That the embryonic second maxillae give rise to the labium has been
shown by Butt (1957) in *Oncopeltus fasciatus* (Dallas), and by other authors
in other insects. Christophers (1960) also believes that the subgena is
the labial area; he homologizes the subgenal and postgenal areas posterior
to the maxillae with the fused bases of the maxillae (cardo and stipes).
He thus believes that in the larval as in the adult stages of mosquitoes
the bases of the maxillae extend to the occipital foramen, forming the
hypostomal area. However, the sclerite which Christophers considers
as the base of the maxilla serves as the origin of pharyngeal, man-
dibular, and maxillary muscles which in most other insects originate
on the tentorium or on the cranial wall (Snodgrass 1935). In the adult
Aedes vexans (Meigen) the maxillary muscles originate on the tentorium
(Peterson Hoyt 1952). On the other hand, none of the postgenal muscles
of the mosquito larva originates on the tentorium. If the larval post-
gena and subgena are to be considered as the fused maxillary cardo and
stipes, then the origins of the various muscles upon them are difficult
to explain. Menees (1958a), studying the embryonic development of
A. quadrimaculatus, observed that the median suture on the ventral head
sclerite in this species is the result of incomplete fusion of the postgenae.

Most sutures which are characteristic of the primitive insect
head are absent from the heads of mosquito larvae. Two cleavage lines
extend anteriorly from a short posterior occipital stem (fig 1). These

cleavage lines may be homologous with the frontal sutures and the epicranial suture of other insects. However, Snodgrass (1947, 1958) and DuPorte (1953) state that the frontal arms of this suture follow diverse paths in different insects, and therefore do not define any specific part of the head. For this reason, in this work head sclerites and mouthparts have been named in reference to muscle origins.

Approximately in the center of the frontoclypeus arise the labral and epipharyngeal muscles (fig. 1) which usually originate on the clypeus, and posterior to these are the origins of the pharyngeal muscles which generally occur on the frons. In the head of *Aedes fitchii* (Felt and Young) larva and in all the other mosquito species examined, there is no demarcation between the areas where the different muscles originate. According to DuPorte (1962) in some insects the boundary between the clypeus and frons, in the absence of an epistomal suture, is fixed by the position of the anterior tentorial pits. In the heads of mosquito larvae, however, the epipharyngeal muscle (usually on the clypeus) originates much posterior to the anterior tentorial arms.

The tentorium in the mosquito larva is represented by anterior and posterior arms. The anterior arms originate on the head capsule medial to the antennae, in the same area where the hypopharyngeal bars arise (fig. 1). The long, slender anterior tentorial arms connect to the short posterior arms on the postero-ventral part of the head. There is no tentorial bridge.

On each side of the head a hypopharyngeal bar connects the hypopharynx to the side of the cranium (fig. 1).

The Labrum

The labrum of the larva of *Aedes fitchii* consists of a narrow transverse sclerite dorsally (fig. 1). Ventrally it is composed of a membranous area to which three brushes are attached, one median and two lateral and movable. The median brush is connected to each lateral labral brush and to the distal part of the dorsal labral sclerite by a membrane which has ben variously named. In the larvae of *Lutzia halifaxi* Theobald, Cook (1944b) referred to it as a "pennicular area.. beset with small oval pits arranged in definite rows." Because of its appearance Christophers (1960) called it the tessellated membrane, and this is the name adopted here (fig. 5). However, this name does not describe the membrane accurately in all the larvae that I examined. This is discussed further below.

In both *A. aegypti* , (Shalaby 1957a) and *Aedes fitchii*, two types of hairs are found on the median brush; long thin branched hairs posteriorly, and short stout hairs with serrated distal ends anteriorly. Both types are shorter on the sides of the brush than medially.

The lateral labral brushes are composed of three types of hairs which differ in length, thickness, curvature, and location. The hairs of the first type are simple, relatively short, thin, soft, without definite curvature, and are located postero-laterally, dorsally, and ventromedially overhanging the pharynx (figs. 1, 3). These hairs, which are attached to the tessellated membrane, do not take part in creating a feeding current. Hairs of the second type are long, simple, thin,

slightly curved at their bases and at their distal ends, and are located in the lateral posterior two thirds of the brush (fig. 3). Anterior to them are hairs of type three. Types two and three take an active part in creating currents. The apices of type three hairs are provided with serrations (17 - 20 per hair). The serrations on the lateral type three hairs are smaller and slightly closer to each other than those on the more medial hairs.

Three types of hairs were found in all the browsing species of *Aedes* and *Culiseta* except in *Aedes cinereus* Meigen and *A. canadensis* (Theo). which have only short, simple hairs on their lateral brushes. When the labral brushes are stained with Mallory's the bases of all the hairs stain red. Next above the bases a narrow layer of blue appears across the hairs and above this layer hairs of type one and two stain red to their tips. Hairs of type three stain partly red above the blue portion but they stain blue apically, in their serrated regions. A large proportion of the most median type three hairs stains completely blue above the red bases. In *A. fitchii* and the other *Aedes* larvae, as well as in the browsing *Culiseta* larvae that were examined, the apices of hairs of types one and two are tapered. Also tapered are the apices of all the hairs of the labral brushes of the filter feeders, *Culiseta morsitans* (Theo.) and *Culex territans* Walker. In the brushes of the filter feeding larvae all the hairs are simple. They all have red-staining bases, blue-staining portions above the bases, and red-staining middle and apical portions. In the filter feeding larvae a large group of hairs, originating medially on each lateral labral brush, overhangs ventrally, partly covering the epipharynx. A smaller number of simple hairs extends in this position in the browsing larvae (fig. 1). In all the larvae that were examined these hairs are red - staining. In the larvae of *Chaoborus americanus* the labral brushes consist of a few hard, short, brown bristles on the small sclerite. In the larva of a *Chironomus* species examined a few labral bristles are red-staining and the remainder are blue-staining. Thus the staining reaction of the labral brushes of the filter feeding and browsing larvae indicates that their hair bases are elastic and the portions above the bases are flexible. Flexibility of these hairs was seen when larvae were observed feeding and also when the hairs were deflected with a needle.

In the mosquito larvae examined all the hairs of the lateral brushes except type one are attached to sclerotized rods which extend transversely across the basal area of the brush (figs. 3 and 4). Salem (1931) seems to be referring to these rods in *Aedes fasciata* (Fab.) (*A. aegypti* L.) when he states that the chitin of the labral brush "exhibits a peculiar striated appearance." Christopher's term for these rods, "cross bars," is used here. On each lateral labral brush of *A. fitchii* larvae between forty-five and fifty of these bars are present and each bears approximately twenty hairs. Thus each lateral brush contains nearly a thousand hairs. A similar number of hairs is present in each lateral brush of *C. inornata* larvae.

The cross bars are cuticular thickenings of the tessellated membrane (fig. 5) with their dorsal ends free in this membrane next to the dorsal sclerite of the labrum. When the cross bars are torn away from the tessellated membrane and the hairs, depressions on them where the
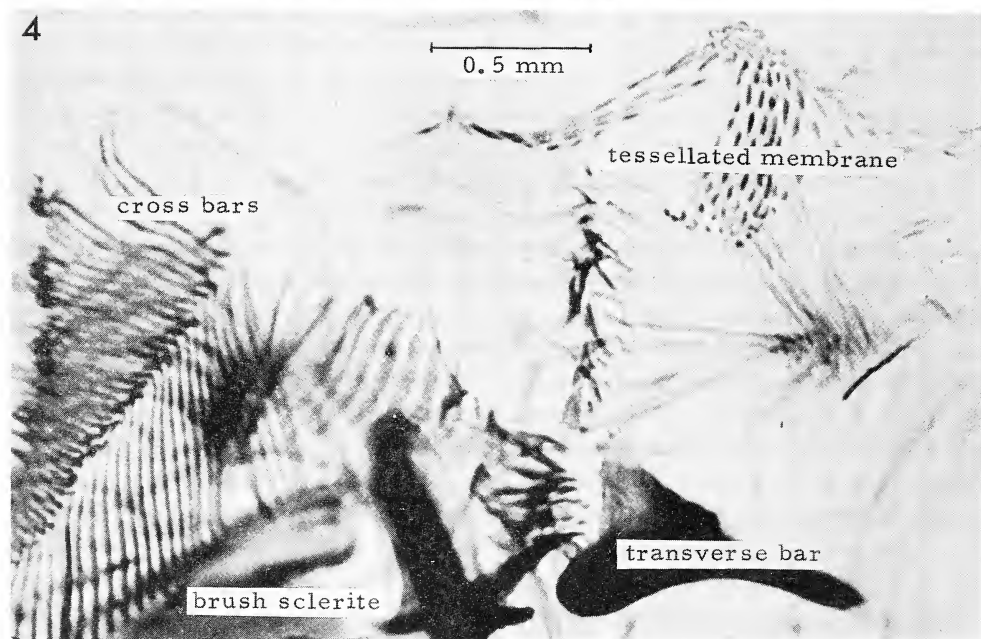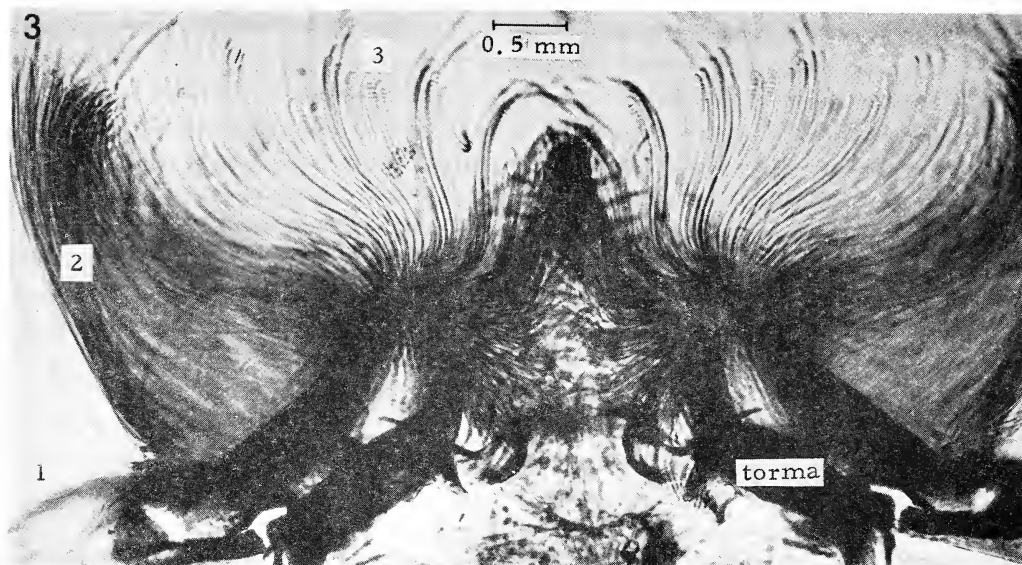
Fig. 3. Ventral view of the labrum of the larva of *Aedes fitchii* with the lateral labral brushes extended. Numbers indicate hair types.

Fig. 4. Details of labral hair attachments of the larva of *Culex territans*.
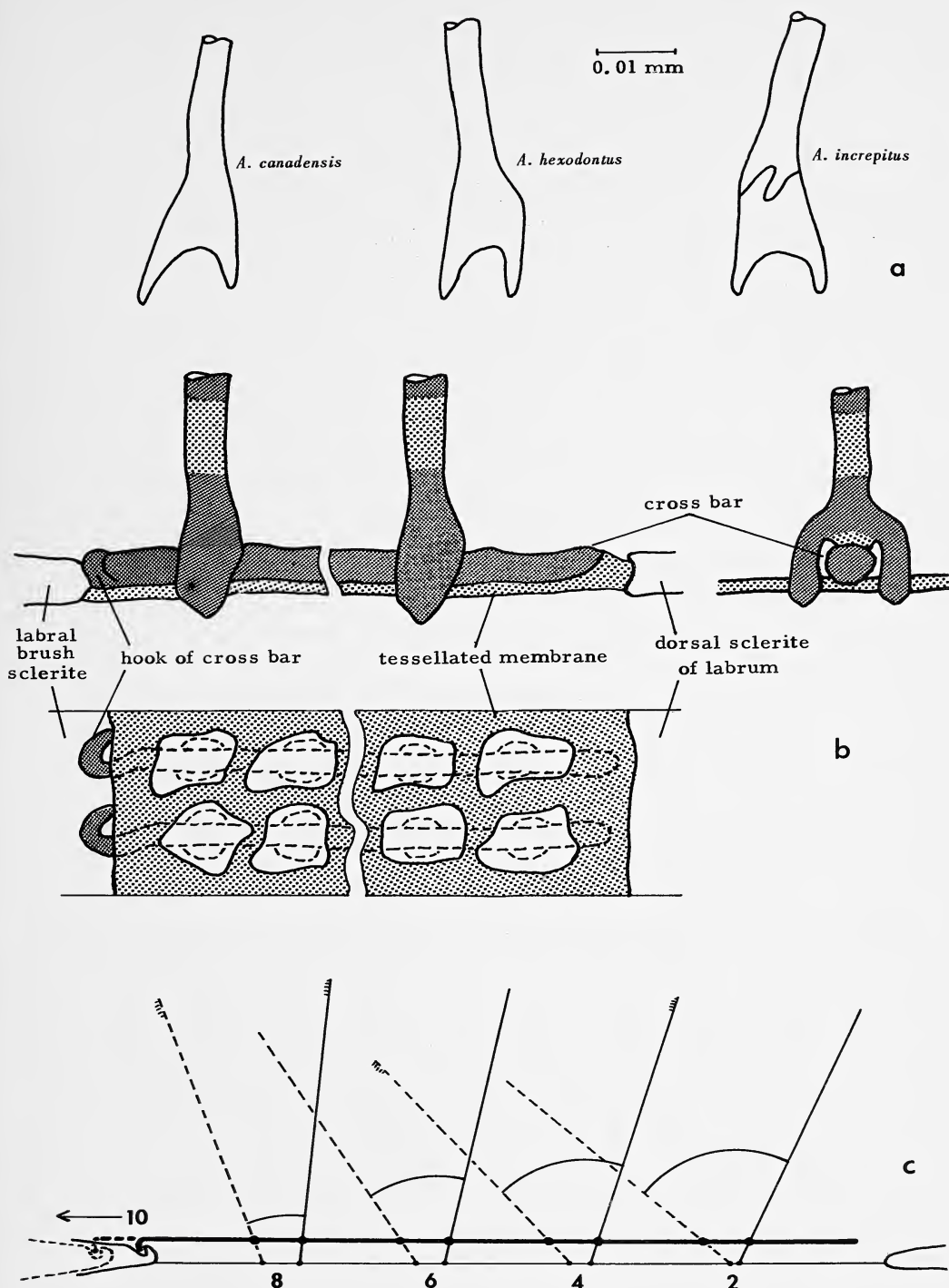
Fig. 5. (a) Forked bases of labral hairs of *Aedes* larvae; anterior views. (b) The relationship between hair base, cross bar, and the tessellated membrane, and the holes and depressions left in this by the removal of hairs and cross bars. Open stipple stretchable cuticle (stains blue); close stipple, flexible but relatively non-stretchable cuticle (stains red). (c) Diagram showing how the hairs are brought together by the increasing angle of movement at greater distances from the brush sclerite, because of differential stretching between the cross bars and the tessellated membrane.

hairs were attached can be seen. The other end of each cross bar is curved into a hook; it terminates in the brush sclerite which is roughly triangular and is attached to the median part of the torma by an apodeme (fig. 3). Muscles that move this sclerite are inserted on the posterior tormal apodeme (fig. 1). When the hairs are pulled off the membrane, their forked bases, the cross bars, and part of the membrane comes with them. This leaves holes in the membrane and confirms that the cross bars are more strongly attached to the hair bases than to the membrane. The hole may be rhomboid, square, pentagonal, hexagonal, oval, or roughly circular and form a mosaic pattern on the membrane which gives it its names. The cross bars leave depressions in the tessellated membrane.

When this complex is stained with Mallory's the cross bars and the hair bases stain red indicating rigidity, while the tessellated membrane and small parts of the hairs above their bases stain blue, indicating stretchability. The edges of the holes may be outlined in red perhaps because of some change in the character of the material of the membrane resulting from tearing.

The ends of the epipharyngeal bar are attached to the posterior parts of both tormae (figs. 1, 3). At the anterior end of each torma a narrow sclerite projects medially. These sclerites are known as transverse bars (Shalaby 1957a) or palatal bars (Christophers 1960). Their structure in *A. fitchii* is slightly different from that in *A. aegypti* as described by the above authors. The bars of *A. aegypti* are slender and from each a small curved sclerite projects anteriorly. In *A. fitchii* they are stout and curved medially, and are attached by thin sclerites to the tormae. In *Culex territans* the bars are straight and have wide basal parts.

In the species examined only the posterior apices of the tormae stain blue; the remainder of these structures with their apodemes retain their brown color. Thus the tormae and their apodemes are rigid, highly sclerotized structures. The associated membranes stain light blue.

The labrum of the predatory *Chaoborus americanus* larva is greatly reduced; it lacks brushes but possesses a few short stiff bristles at the tip of the labral sclerite (Cook 1956). These bristles stain dark red.

The Epipharynx and Preoral Cavity

The epipharyngeal apparatus lies between the posterior ends of the tormae and combs food particles from brushes to the mandibles. Schremmer (1949) called it the "Epipharynx-apparat" because it is musculated and has an active rather than a passive function.

The structure of the epipharynx in the species examined is very similar to that described by Shalaby (1957) and Christophers (1960) in *A. aegypti*. In *A. fitchii* and the other browsers the hairs are coarser than in *Culiseta morsitans* and *Culex territans*. The spines and hairs stain dark red in *A. fitchii* which indicates medium hardness; they stain lighter red in *C. morsitans* and *C. territans* and are probably softer in these species. The epipharyngeal bar stains medium blue in all specimens. That this flexible structure can move anteriorly and posteriorly has been observed in living larvae of *A. aegypti* and *C. territans*

The post-epipharyngeal area consists of a membrane between the epipharynx and the pharynx. It is similar to that described by Cook (1944b) in *Theobaldia incidens* (= *Culiseta incidens*). Two pairs of muscle strands originate on the frontoclypeus, one of these forks before its insertion in the membrane between the epipharynx and the pharynx. Since these muscle strands have a common origin on the cranium medially of the antenna (fig 1), I consider them as fascicles of one muscle, the postepipharyngeal.

The Mandibles

The mandibles of mature *Aedes fitchii* larvae consist of flattened, roughly quadrilateral lobes with their mesal ends produced into strongly sclerotized toothed processes and lower seta-bearing lobes. They are similar to the mandibles of most culicine larvae which have been described by other authors.

On the mesal margin of each mandible is found a fringe of pigmented, long, mesally directed setae with stout bases and sharp points. Shalaby (1957a) called this fringe the mandibular comb when he described it in *A. aegypti* . The number of the curved, stout and sharply pointed setae varies in fourth instar larvae of the species that I examined. Eleven were usually found in *A. fitchii* , nine in *C. inornata* , and fifteen in *A. aegypti* . Another series of setae extends meso-dorsally from the dorsal side of the mandible, medially of the large lateral bristles; this series Shalaby names the mandibular brush. In *C. inornata* it usually consists of 40 setae; in *A. fitchii* of 54. The number of lateral bristles is variable between species, but constant in all the species seen; in *A. fitchii* two are present and in *C. inornata* three. When the mandibular brush and comb setae of the *Aedes* and the *Culiseta* browsing species are stained with Mallory's their bases stain blue, and thus are soft; the remaining parts stain dark red, and are harder. The mandibular setae of the filter-feeding species , *Culiseta morsitans* and *Culex territans* are softer than those of the browsing species. The lateral bristles remain brown in all the species examined. All the mandibular bristles and setae in the mandible of *Chaoborus americanus* stain dark red or remain brown.

The number of teeth in *A. aegypti* , as described by Shalaby, is similar to that in *A. fitchii* and to the other *Aedes* species that were examined. The number of ventral teeth in *C. inornata* is similar to that found in the browsing *Aedes* species, but dorsally only three teeth are present in *C. inornata* whereas five are present in all specimens of all the *Aedes* species. The extent of heavy sclerotization in the tips of the mandibles, mainly the teeth, is approximately the same in *C. inornata* and the browsing *Aedes* species. The heavily sclerotized area is smaller in the filter feeders, and it is largely extended in the predatory *Chaoborus americanus* and *Mochlonyx velutinus* . These characteristics agree with the characteristics of browsers, filter feeders, and predators that Surtees (1959) discusses. Medially, on the dorsoventral ridge of the mandible a group of long spines reaches the anterior part of the pharynx. Schremmer (1949) discusses the function of similar spines on the mandible of *Anopheles maculipennis*. Anterior and posterior mandibular articulations are indicated in fig. 1.

The Maxillae

Each maxilla of *A. fitchii* (fig. 7) consists of a rectangular flattened lobe which bears a brush of long hairs apically, and a series of three rows of short hairs medially in an area demarcated by a suture on the oral (dorsal) side. Proximal to the palpus is a triangular sclerite about half the width of the main lobe, which is attached to these structures and to the post gena by a membrane. This sclerite bears a spine medially. Baso-ventrally the maxillary palpus bears sclerotized processes which articulate with a postgenal articular process inside the head (fig. 1). The mandible also articulates with the postgena and the maxilla at this point. Two muscles are inserted in the center of the main maxillary lobe; a single strand originates on the subgena mesally to the posterior tentorial pit, and a double strand originates on the postgena posterior to the eye (fig. 1).

To decide what parts of the maxilla of *A. fitchii* larvae are homologous with parts of maxilla of other insects, the relation between sclerites and musculature must be considered. It is generally accepted that as Imms (1944) states "... the Mecoptera are the nearest living representatives of ancestors of Diptera..." This view is also expressed by Applegarth (1939), Ferris and Rees (1939), Potter (1948), Hinton (1958), and others. We should therefore look for homologies of the maxilla of the mosquito larva in the Mecoptera and in other members of the suborder Nematocera. The palpus is the only structure on the homology of which all the authors agree. Since the palpus is connected to the base of the main maxillary lobe, and since the palpus in all insects is connected to the stipes, it seems logical to consider this lobe as the stipes. According to Snodgrass (1936) and Das (1937) the stipes can be distinguished by the origin of the muscles of the palpus. However, this criterion does not apply when the palpal muscles are absent as from mosquito larvae and larvae of *Tipula* and *Bibio* as described by Das (1937) and Cook (1944a). The two muscles that are present in this structure are probably the cranial flexors of the stipes (rather than of the lacinia). The double strand which originates on the postgena is one of these, and the adductor of the stipes which usually originates on the tentorium is the other. In the culicid larva the origin of the latter has shifted to the subgena.

Snodgrass (1935) and Das (1937) hold that the lacinia has a cranial flexor and the galea has only a stipital flexor in larval and adult stages of many insects. Das also states that many larvae lack the flexor of the galea, but when the lacinia is present its cranial flexor is always retained. The same author adds that the cranial flexor of the lacinia plays an important role in the interpretation of the lobes. No trace of stipital flexor was found in any culicid larva examined. The only cranial flexor present is inserted so close to the median side of the main lobe that it is almost on the bristle-covered area which is demarcated by a suture on the oral side of the lobe (fig. 7). Furthermore, this median bristly area functions as a lacinia. Therefore I agree with Shalaby (1957a, 1958) that this part of the maxilla is the lacinia, and that the cranial flexor of the lacinia now functions as a stipital flexor.

In the larvae of *Panorpa* both galea and lacinia are present (Das 1937); in *Apterobittacus* only the lacinia is present in the larval stage and the galea appears in the pupal stage (Applegarth 1939); in both *Tipula* (Das 1937) and *Bibio* (Cook 1944b) only the lacinia is present in the larval stage. The
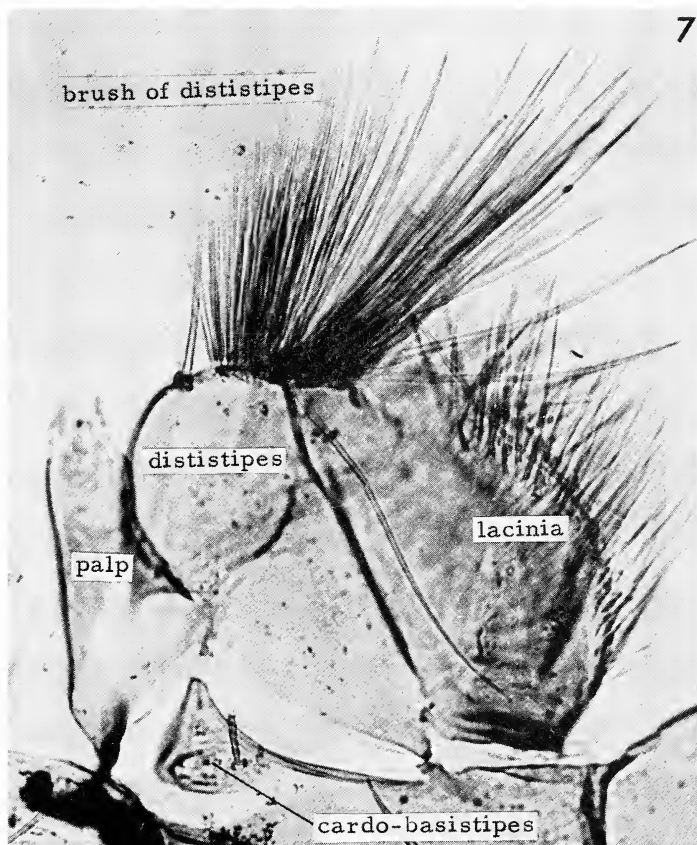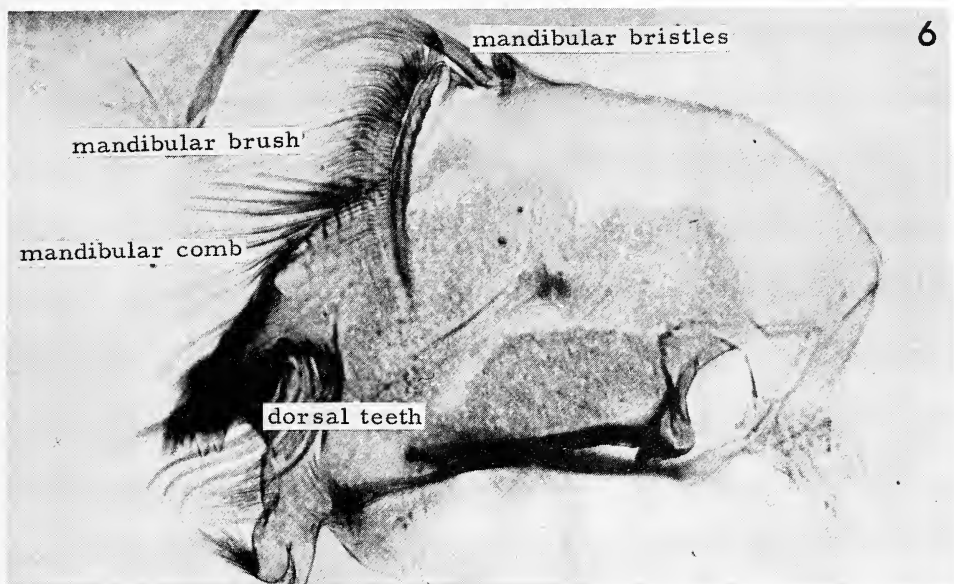
Fig. 6. Ventral view of the left mandible of mature larva of *Aedes fitchii*

Fig. 7. Dorsal view of the left maxilla of mature larva of *Aedes fitchii* .

triangular sclerite which is considered as the palpifer by most authors
I believe to be at least a partial vestige of the cardo. In the larva of
Panorpa the cardo has a relative size, shape and position similar to that in
the mosquito larva, and it also lacks musculature (Das 1937). In the lar-
vae of each of *Apterobittacus*, *Bibio*, and *Tipula* species the structure named
as cardo by the respective authors, is proportionately larger than in the
larvae of *Aedes*, *Culex*, and *Culiseta*. In the former three larvae the so-
called cardo extends posterior to the stipes and the palp. If this structure
is homologous with the triangular sclerite in the mosquito larva then this
sclerite must be the cardo and not the palpifer. However Hinton (1958)
points out that the stipes is divided into a basistipes and dististipes in all
the Panorpoidea except the more specialized Diptera. The same author
further states that failure to recognize the fact that the stipes is sub-
divided in primitive forms of all recent orders of the Panorpoidea has
resulted in the misidentification of the dististipes as the palpifer. Hinton
also states: "in the Panorpoidea in which the cardo has become fused to
the basistipes the combined structure which may be called the cardostipes
has almost without exception been identified as the cardo and the disti-
stipes as the stipes. For instance, the cardo plus basistipes of *Bibio* is
called the cardo and the dististipes is called the stipes by Imms (1944) and
Cook (1949)..." In the light of Hinton's statements then I consider the
triangular sclerite of the mosquito larval maxilla as homologous with
the fused cardo and basistipes. The main maxillary lobe is the disti-
stipes plus the lacinia. In addition Hinton mentions that within the
Nematocera a fusion of the cardostipes with the dististipes takes place for
example in the Culicidae, but he does not specify in what group of the
Culicidae. He may be referring to the genus *Anopheles*, for in that genus
there is no triangular sclerite proximal to the maxillary palp and the
dististipes as in the genera *Aedes*, *Culex*, and

Essentially the same structural arrangement of the maxilla
was found in all the *Aedes*, *Culex*, and some *Culiseta* larvae that I
examined. Some difference from the browsers was found in the shape
of the maxillae of *Culex territans*, *Culiseta morsitans*, *Aedes canadensis*, and
A. cinereus Each maxilla in these species is cone-shaped, wide at the
base and narrow at the apex where a brush of simple hairs is attached.
The maxillae of most browsers are similar in shape to those of
Aedes fitchii. Between the browsers and filter feeders differences occur
in the number and length of hairs on the distal end of the dististipes and
on the lacinia. In the maxillae of both filter feeders and browsers the
apical brush hairs of the dististipes are longer than the lateral hairs of
the lacinia, and in the filter feeders all these hairs are proportionately
longer than in the browsers. The longest maxillary hairs in *Culex territans*
and *Culiseta morsitans* are approximately one and a half times as long as
the dististipes; whereas the homologous hairs in *A. fitchii* and the other
Aedes browsers are only approximately as long as the dististipes, and
in both *Culiseta inornata* and *C. impatiens* (Walker) they are half the length of
the dististipes. The maxillary brushes of the browsing *Aedes* species are
composed of more hairs than those of the filter feeding species. The
maxillae of *C. inornata* and *C. impatiens* larvae have brushes consisting
of very few hairs, thus resembling the maxillae of predatory larvae.

Another similarity of the maxillae of these two *Culiseta* species to the predatory larval maxillae is the fusion of the palps with the cardobasistipites.

With Mallory's stain the bases of the maxillary brush hairs of browsing larvae stain blue and the remaining parts red, but the whole hairs stain blue in filter feeders. Thus the maxillary brushes of browsers are stiff, a feature of obvious value in their activity.

The short medial bristles of the lacinia are arranged in three rows in all the species that I studied; they are more numerous in browsers than in filter feeders. These hairs are longer in *A. fitchii* and the other *Aedes* browsers than in *C. inornata* and *C. impatiens*. In all the browsers these hairs stain red, indicating moderate stiffness. The hairs of the lacinia of the filter feeders stain blue and thus are soft.

The Labium and Hypopharynx

I consider the labium of the larva of *A. fitchii* to consist of the prementum and the submentum. This view is in agreement with Cook's (1944b) interpretation for other genera. The prementum (fig. 2) is a rectangular membranous area bearing a series of serrated sclerites and papillae, and is situated between the hypopharynx and the mouth opening dorsally, and the triangular serrated submental plate ventrally.

Dorso-ventrally two long sclerites extend through the centre of the prementum and dorsally terminate ventral to six small serrated sclerites which project ventrally from the membranous base. On the sides of the membrane three serrated plates are situated ventrally. These three plates are connected to each other, and dorsally to the small central serrated sclerites. Each plate has a different number of serrations, which vary in different species. In *A. fitchii* the dorsal plate has four serrations, the median plate nine, and the ventral plate five. Six larvae of each of two closely related species, *Aedes hexodontus* and *A. punctor* were also examined, and the average numbers of serrations were found to be: dorsal plate 5 serrations in *A. hexodontus*, 4 in *A. punctor*; median plate 6 in *A. hexodontus*, 9 in *A. punctor*; ventral plate 6 in *A. hexodontus*, 10 in *A. punctor*. This may be a useful taxonomic character for separating closely related species. Considerable care is required in preparing the slides if the serrated plates are to be seen clearly.

Since these plates in all the species of *Aedes*, *Culiseta*, and *Culex*, that were examined stain light red basally and dark red to orange distally, they are quite hard. This is understandable because the mandibular teeth which are of similar hardness strike against them. The hardness of both structures could be felt with dissecting needles. In the *Aedes* species a group of broad, apically serrated hairs originates on the mid-ventral side of the premental lobe. Broad, but not serrated hairs occur in the same position in the *Culiseta* and *Culex* species. These hairs are numerous in *Aedes* and *Culiseta* but very scarce in *Culex*. In all the species examined they stained medium red with Mallory's.

On the premental lobe laterally, between the central and the lateral serrated plates four small papillae are present on each side in all the species of *Aedes*, *Culiseta*, and *Culex* that I examined. The most

posterior papillae are double on each side; the more anterior two arise singly. Two similar papilla-like processes are present in the membrane dorsally between the serrated plates and the salivary duct opening. In all the species considered the papillary structures stained red, and the basal membranes light blue. In feeding larvae, food often collected in the spaces between the papillae and the serrated plates.

It is difficult to homologize the structures of the labium because of its degenerate nature, but since a pair of muscles attaches the rectangular lobe to the subgena medially to the posterior tentorial pits (fig. 1), these muscles are considered as the premental muscles by Cook (1944b, 1949), Snodgrass (1959), and others, Snodgrass refers to the lobe as the labial plate. I agree with Cook in calling it the prementum.

The premental membrane is dorsally suspended from the hypopharyngeal bars. A weak suture continues between these bars and dorsally of the premental membrane, thus demarcating an oval membranous hypopharyngeal area above the prementum. The opening of the salivary duct is located between the premental and hypopharyngeal lobes. This was so in all the species examined including *A. aegypti* although Christophers (1960) shows it in the center of the prementum.

The triangular serrated sclerite below the prementum has been variously named (table 1). I agree with Cook (1944b, 1949) that it represents the submentum. Salem (1931) considered it homologous with the submentum, but thought that the customary name, mentum, should be retained. Snodgrass (1959) believed it to be an extension of the subgena. Jones (1960), following Snodgrass, calls it the hypostomium in the larva of *Anopheles quadrimaculatus* . My main reason for disagreeing is that in all the species examined this sclerite articulates with the subgena, and therefore is unlikely to be an extension of it. Generally the submentum of insects articulates with the ventral part of the cranium (Snodgrass 1933). Snodgrass (1959) however, does not mention that this triangular structure arcticulates with the subgena. He states that it is continuous with the subgena, as in the head of *Chironomus* described by Grouin (1959) who calls it the hypochilum. Miall and Hammond (1891) indicate that this plate in *Chironomus* seems to correspond to the submentum of orthopterous insects.

The submentum stains orange basally with Mallory's and remains dark brown apically in all the *Aedes* , *Culex* , and *Culiseta* larvae I examined. It is thus a very hard structure. In the species examined the number of serrations on it in mature larvae is usually constant; data are given in table 2.

The lightly sclerotized fringe of hairs (figs. 1, 2) attached to the submentum ventrally stains similarly; I consider it a part of the submentum since it is very intimately connected with this structure. Cook (1944b) calls it the aulaeum.

The Pharynx

The structure and musculature of the pharynx of *A. fitchii* and *C. inornata* larvae are similar to those of *Theobaldia incidens* (= *Culiseta incidens*) described by Cook (1944b). The large dorsal and ventral sclerites stain light orange in all the *Aedes* , *Culex* , and *Culiseta* larvae

TABLE 2 - The numbers of serrations on the submentum of the larvae
of mosquito species.

Species	No. of submental serrations		Species	No. of submental serrations	
Aedes spp.			*Aedes* spp.		
campestris	27.0	(1)*	*sticticus*	21.6±0.5	(3)
canadensis	20.0	(1)	*stimulans*	28.0	(1)
cinereus	25.0	(1)	*vexans*	26.0±0.7	(5)
excrucians	20.5	(2)	*Culiseta* spp.		
fitchii	20.6±0.9	(20)	*impatiens*	25.0	(1)
hexodontus	24.6±1.1	(5)	*incidens*	18.0	(1)
implicatus	18.0	(1)	*inornata*	23.9±2.2	(17)
increpitus	25.0	(1)	*morsitans*	19.0	(2)
impiger	20.5±1.3	(4)	*Culex* spp.		
pionips	24.0	(2)	*pipiens*	21.0	(2)
punctor	27.1±0.7	(6)	*tarsalis*	13.0	(2)
riparius	23.0±0.7	(5)	*territans*	13.0	(2)

* average ± S. D. of the mean (where applicable);
number of specimens examined in parentheses.

examined. The lateral dorsal hairs stain light red, and the inner filtering hairs stain light blue in most species. Schremmer (1949) described the filtering function of the pharyngeal hairs in *Anopheles maculipennis* larva.

Discussion

It is difficult to decide on the homologies of degenerate structures like the maxilla and labium of mosquito larvae. Shalaby's (1957 d) interpretation of the triangular labial sclerite as the paraglossa, and the aulaeum as the glossa is unique, and seems unreasonable. The areas which I consider as the hypopharynx and the prementum Shalaby regards as the hypopharynx. Medio-laterally on the premental lobe a pair of muscles is inserted. These muscles originate on the ventral sclerite of the head which Shalaby considers as the submentum and which I regard as the subgena. It is difficult to agree with Shalaby's interpretation of the labium and the hypopharynx for the following reasons: firstly, as far as is known, the hypopharynx in insects is not connected with the paraglossa, but in the mosquito larva, in Shalaby's interpretation the " hypopharynx " is firmly attached to the "paraglossa". Secondly, other authorities on the morphology of insect larvae (Cook 1944, 1949; Hinton 1958) state that the retractor muscles of the hypopharynx are absent in Diptera. Thirdly, when the retractors of the hypopharynx are present they arise on the postoccipital ridge in the Trichoptera, and on the tentorial bridge in the Lepidoptera (Hinton 1958), but not on the "submentum" where these muscles originate in the mosquito larva according to Shalaby's interpretation.

Very few muscles which could serve as guides to homology are present, and this is partly why disagreements exist among the various morphologists who have studied mosquito larval mouthparts. Ferris (1948) postulates the following principle: "..... the evolutionary changes are first to be accounted for by modifications of pre-existing structures, or by loss of pre-existing structures;..... Only after these possibilities have been exhausted will we assume that a completely new structure has been developed. ..." This principle can be applied to mosquito larvae and to the larvae of other primitive Nematocera when we compare them with panorpoid larvae. In mosquito larvae noticeable modification from *Panorpa* is seen in the labrum and in the mandibular teeth. Losses and fusions of pre-existing structures are evident in the mosquito larval maxilla and the labium.

A difference was found in the hardness and flexibility of the cuticle of the mouthparts of the filter feeding, browsing, and predatory mosquito larvae. Essentially, the mouthparts of the filter feeders are rather soft except for the labral brush hairs and the mandibular teeth; the mouthparts of the browsers are harder, and the mouthparts of the predatory larvae are the hardest of all, especially the mandibles, which are highly sclerotized.

The tips of the simple labral brush hairs of the filter feeding and browsing larvae are softer than the main parts of the hairs. The labral brush hairs of these groups of larvae are much harder than they appear to be since they are refractory to stain until after boiling in a relatively strong (8%) solution of KOH. It was interesting to find that

the serrated ends of the lateral labral brush hairs of the browsing larvae stain blue and thus are soft combs rather than hard ones as they might be expected to be when their function is considered. Since they are soft it is probable that when they rub over surfaces soft particles are detached and then directed towards the mouth. The physical characteristics of the cuticle were estimated by manipulating the mouthparts, and the impressions obtained agreed with the indications from staining.

The serial row attachment of the labral brush hairs to their respective bars is similar in the browsing and the filter feeding larvae. Christophers (1960) also noted that the hair attachment is similar in the larvae of a *Culex* species and of *A. aegypti* .

In table 3 it is indicated that a reduction occurs in the numbers of hairs or bristles on the various mouthparts from the filter feeders to the predators. In the same series an increase in the sclerotization of the mandibular teeth is evident.

TABLE 3 - Similarities and differences in the mouthparts of filter feeding, browsing, and predatory mosquito larvae.

	Labral brush hairs	Maxillary hairs	Premental hairs		Mandible	
				Sclerotization	Sclerotization	Plane of action
Filter feeders						
C. morsitans	many long thin simple	many very long	few short	moderate	slight	
Intermed.						
Aedes cinereus	many thin simple	very many long	many long	moderate	heavy	nearly dorso-ventral
Browsers						
Aedes fitchii	many thick serrated	very many long	many long	moderate	heavy	
Culiseta inornata	many thick serrated	few short	many long	moderate	heavy	
Predators						
Mochlonyx velutinus	few short thick serrated	very few very short	many mostly long	slight	very heavy	antero-lateral
Chaoborus americanus	very few thick short serrated	none	very few short	none	very heavy	

It is interesting to note that the same genus is represented by filter feeding (*Culiseta morsitans*) and browsing larvae (*C. inornata* and *C. impatiens*) whose mouthparts tend towards the predatory type. Most

of the *Aedes* species that were studied are browsers, but the larvae of *Aedes cinereus* Meig. and *A. canadensis* lack serrations on their labral brushes, have more weakly sclerotized mandibular teeth than the other *Aedes* species, and their maxillae are similar to those of the filter feeders. Thus morphologically these species seem to be intermediate between the filter feeders and the browsers.

From table 3 it is also evident that the plane of action of the mandibles in the predatory larvae tends towards that of the longitudinal axis of the body which is a character common both among the larvae of the higher flies, according to Cook (1949), and among predators generally.

FUNCTION OF THE MOUTHPARTS OF MOSQUITO LARVAE

Procedures

The movements of the mouthparts of mosquito larvae and actions resulting from these movements were studied in two situations: behaviour of larvae (mostly *Aedes*) was observed in the muskeg pools in the Flatbush area (100 miles north of Edmonton) in the summers of 1960 and 1961; more extensive observations were made on active larvae in artificial containers in the laboratory.

After being collected the larvae were kept in pint glass jars, and in order to retard their development when not being observed they were kept in the refrigerator at 40°F. The larvae were observed in groups and individually in the glass jars and some details of movements of their mouthparts were seen with the aid of a 10X hand lens. Individual larvae were placed in small vials and their mouthparts were observed from the side with a hand lens. A viscous solution of an inert material such as methyl cellulose was also used to slow down the motions of the mouthparts so that details of their actions could be studied.

Larvae of *A. aegypti* and *Culiseta inornata* reared in the laboratory were observed. Other species of *Aedes* and *Culiseta* were collected in the areas of Flatbush, Edmonton, Lake Hastings, Banff, and Seebe, Alberta. The larvae were identified with the keys of Rempel (1953) and Carpenter and La Casse (1955).

Since the mouthparts are ventral it was desirable to observe larvae from the ventral side; three methods were used for this. For all the methods a container was made by cutting a 1 in long piece of a plastic vial of 1 in diameter, and gluing it to a microscope slide which formed the bottom. The container was filled with either pond water or distilled water and food was added. Usually one larva was studied at a time, but sometimes two were observed in the same dish.

By means of two concave mirrors, light from two microscope lamps was directed on the larva through the bottom of the container. An image of the ventral surface of the larva was reflected by two plane mirrors at 45°, one below the container and one below the objective of a stereo-binocular microscope. A satisfactory view of the mouthpart

movements was obtained in this apparatus. The movements were most clearly seen at magnifications of six or twelve diameters. More detail was seen under 25X and 50X, but the images were blurred, especially at 50X.

A second method of observing the mouthparts was by turning the body and eyepiece of the binocular microscope upside down and focussing on the larva above the microscope. The best image was obtained by this method which was used most often. Fluorescent light from above and focussed light from below were used separately and in combination.

A third and most convenient method of observing the movements of larval mouthparts was through a metallurgical binocular microscope with the stage above the objective lens. In this method it was possible to have the light coming only from above.

Particles of activated charcoal or methyl red were placed in the containers with the larvae to show the directions of the currents set up by the mouthparts.

Observation of the Mouthparts in Action

The operation of the lateral labral brushes was studied by direct observation of living larvae and by manipulation of prepared material. The mechanism of action in each type of mouthpart is described separately below.

Browsers

In this group contraction of the labral muscles exerts tension on the brush sclerite which in turn pulls the tessellated membrane and the cross bars by their hooks. This causes the hairs of the brush to move ventro-medially. The hairs spring back outwardly through the elasticity of the tessellated membrane. The inward and outward movement of the hairs is thus caused by the differential elasticity of the tessellated membrane and the cross bars. The bases of the hairs are connected with the cross bars, and fork on either side of them (fig. 5). The bifurcations are short, and their ends terminate in the tessellated membrane below the cross bars. The stretch of the tessellated membrane allows the part of the hair which is attached to the rigid cross bar to move more than the tips of the fork, so that the hair pivots about this attachment to the cross bar, and its tip swings ventro-medially. Relaxation of the labral muscles allows the hairs to return to their original positions through the elasticity of the tessellated membrane.

The angle through which a hair swings should increase with its distance from the brush sclerite since it is separated from this by a greater length of the elastic membrane. This would have the effect of bunching the hairs together in the median position and allowing them to fan out in the lateral position, which was repeatedly observed to happen.

The main feeding current, produced by the lateral labral brushes, is directed toward the epipharynx and the mouth by the median labral brushes. When creating a current the lateral labral brushes vibrate from

TABLE 4 - Mean frequency and duration of movements of the lateral
labral brushes of larvae over one minute periods at 24 to
27°C.

Time in min.	4th instar means of 3 larvae		2nd and 3rd instars means of 4 larvae		4th instar means of 3 larvae	
	Cycles per sec.	Average duration sec.	Cycles per sec.	Average duration sec.	Cycles per sec.	Average duration sec.
5th	1.7	28.5	2.6	11.0	2.0	30.0
10th	2.7	17.7	3.0	11.0	3.3	13.0
15th	3.3	37.0	2.4	62.5	1.8	8.0
20th	3.1	35.5	3.1	24.0	1.8	7.0
25th	3.4	65.0	2.8	16.4	1.8	7.0
30th	3.5	27.0	2.6	31.0	1.8	4.0
35th	4.0	25.0	3.0	11.7		
40th	3.7	36.4	2.8	12.0		
45th	3.6	68.0	2.1	6.0		
50th	4.5	32.5	4.6	13.7		
55th	3.7	61.0	3.2	12.5		
60th	3.5	40.3	3.4	16.6		

postero-medially to antero-laterally.   The brushes of *A. aegypti* may
vibrate for as long as 2.5 min without stopping.   Then they usually stop
for 5 - 10 sec before resuming.   The more usual timing is vibration for
50 sec, stop for 5-10 sec. and then vibration again.   In *Culiseta inornata*
and in the browsing *Aedes* species the duration of movement is shorter.
Frequency and duration of movements for *C. inornata* and *A. aegypti* and
indicated in table 4.   Table 5 shows activity of individual 4th instar
C. inornata larvae, each of which was observed for 30 minutes.   During
each 30 minute period the activity of the whole body and of the mouthparts.
was observed, and the percentage of time spent in each observable activity
was calculated.

Feeding and locomotory activities of approximately 50 *C. inornata*
larvae were observed individually for various periods of time throughout
the period of the study, and many more were observed in group behaviour.
Much similarity was noticed in the pattern of behaviour of the various
individuals, and almost any larva could be chosen to represent the common
sequence of activities. The following is a summary of the activities of a
4th instar *C. inornata* actively browsing larva (no. 6 in table 5), observed
for 20 min at a magnification of 25X.   The container was filled with
pond water.

During the first minute the larva was stationary; it was suspended from the water-surface with labral brushes extended. For the next 10 sec the labral brushes, the maxillae, and the mandibles created a current, then a 10 sec period of rest followed with the mouthparts retracted and the whole body still. During the first 5 min period such a succession of currents in which all the mouthparts participated was produced four times, and each time the labral brushes moved about 15 times. The mandibles and the maxillary brushes also moved approximately as many times as the labral brushes.

TABLE 5 - Percentage of time spent by 4th instar *C. inornata* larvae in different activity states over a 30 min period.

Body of larva		Labral brushes		
Stationary	Moving	Retracted	Extended	Moving
48	**52**	37	48	15
52	48	18	59	23
62	38	22	57	21
44	56	0	63	37
53	47	6	25	69
12	88	35	29	36
45	55	5	16	79

The larva browsed on a filamentous piece of plant for ten seconds. The piece of plant was enclosed by the labral brushes and the mandibular teeth struck it. Then the larva moved to a chickweed leaf and browsed on its edges for 18 sec. The median hairs (type 3) of the lateral labral brushes held the edges of the tissue while the more lateral hairs (type 3) of the brushes produced a current which moved the larva forward along the leaf. The mandibular teeth struck the tissue. Then the tissue was left and further currents were produced by the mouthparts. Pieces of debris passed into the current which was produced continuously for approximately 20 sec. Mandibular teeth chopped off small pieces of decayed material, some of which went into the mouth and the remainder moved out with the current. Again a piece of plant tissue was browsed upon, and was then propelled posteriorly. When one end of the plant was at the submentum the aulaeum clung to it for a few seconds, but with the subsequent current the tissue was forced posteriorly and towards the bottom of the container.

During the next ten minutes continuous movements of the mouthparts occurred 15 times, each time the duration of the current was approximately 15 sec.

The amount of brush movement and body movement varies among larvae of different ages and different species. Fourth instar larvae are more sluggish than younger ones, and 4th instar *Culiseta inornata* and *Aedes fitchii* larvae are more sluggish than the corresponding instars

of *A. aegypti* (table 4). Shannon (1931) and Christophers (1960) also noticed that *A. aegypti* larvae moved considerably faster than the larvae of most other species of mosquitoes. Fourth instar *A. aegypti* larvae can ingest charcoal particles faster than 4th instar *C. inornata* larvae. When activated charcoal was placed in a container with 3 *A. aegypti* larvae and in another container with 3 *C. inornata* larvae (all 4th instar), the guts of the former were filled in from 90 to 105 min, whereas the guts of the latter species were only filled after 3.5 hr. Larvae of all the species observed moved faster and more frequently when they were stimulated to activity by other organisms (*Daphnia*, *Cyclops* etc.).

When the brushes are not rhythmically beating to create a feeding or a locomotory current they remain extended and separated into rows of four or five layers (fig. 7), or they are retracted (fig. 7). Particles which have been brought close to the mouth by the current continue streaming towards the mouth through the spaces between the rows of hairs, or if the brushes are retracted the particles come to rest on the maxillary brushes. If these are extended the particles stream into the mouth and some settle on the hairs of the pharynx, the mandibles, the maxillae and the prementum. The separation of the labral brush hairs into several rows (fig. 7) is possible because of the basal structure of the brush. Each row of hairs can be moved about the axis of its cross bar. Several rows can move in one direction together, and thus water can flow through the spaces between these groups of hairs. It also seems that the water currents can force the labral brushes to close. The muscles that insert on the tormal apodemes (fig. 3) extend the brushes by contraction. Relaxation of the labral muscles allows the hairs to return to their original positions through the elasticity of the tessellated membrane. This can be demonstrated in preserved specimens. The contraction of these muscles and of the epipharyngeal muscles was observed in living larvae of a filter feeder, *Culiseta morsitans* .

The feeding currents of *Aedes* and *Culiseta* browsers are fast and can carry large as well as small particles. Objects about one third the size of a larval head can be circulated in the stream (fig. 8), the current and the particles reach as far posteriorly as the fourth and fifth abdominal segments and extend about the same distance in front of the larva. Such circulation of particles can be observed when the larva is suspended in water and also when it lies on its dorsal side in an observation cell.

When a larva feeds just above a loose sediment (fig. 8) or browses its way forward through debris in a container, the particles that do not enter the mouth fall to the bottom of the container or cling to the brushes; they do not return to the feeding current. The feeding current is effective only in front of the larva, and it is slowed down behind the larval head. The water flows ventrally rather than posteriorly below the body of the larva. When the larva leaves the browsing area many particles remain on its labral and maxillary brushes, since what does not fall to the bottom of the container sticks to the brushes. Some filtering is done by the labral brushes, especially by the median serrated ends of the lateral labral brush hairs, quite large particles are found clinging to them. Since particles only slightly smaller than these have been found in the pharynx and in the intestine, and since most food seems to come into the mouth via the

labral brush current, it seems reasonable to assume that the particles
which passed into the mouth and eventually into the gut were filtered out
by the brushes. The serrated brush hairs are useful in browsing, for as
they move along surfaces they detach particles from them, many of which
are consumed.

With its labral brushes a browsing larva can attach itself to a grass
stem, to the side of a container, or to a body of a pupa or another larva.
While the labral brushes cling to surfaces the maxillary brushes produce
a current. The browser's maxillary brushes can create currents that are
as strong as those of the labral brushes. This was observed in fourth
instar larvae of the following species:*Aedes cataphylla*Dyar,*A. sticticus*(Meigen),
A. communis (De Geer),*A. fitchii ,A. punctor,A. riparius,A. canadensis* , *Culiseta inornata* and
C. impatiens . The larvae can also browse on parts of their own body,
especially on the posterior regions of the abdomen. This was observed
particularly in containers where *Aedes* and *Culiseta* larvae were crowded.
Many times larvae, especially *C. inornata* and *Aedes canadensis* , were seen
browsing on the tips of their own abdomens and creating currents
at the same time. They were in loop-like positions and moved in
circulating paths of the water surface. This was particularly notice-
able in the laboratory with the larvae of *A. canadensis*; on one occasion in
June 1960, 20 to 30 larvae turned in this manner for several minutes,
individual larvae turning for as long as five to six minutes. Christophers
(1960) states that larvae browse on parts of their own bodies, especially
on the posterior parts, when they are starving. My observations
agree, for in situations where this behaviour took place little food was
present.

Interfacial feeding (Renn 1941, and fig. 8) is a common method of
feeding in the *Anopheles* filter feeding larvae. Third and fourth instar
C. inornata , *A. aegypti* , *A. fitchii* , *A. punctor* , and *A. riparius* larvae also brow-
sed at the water surface without browsing on their siphons at the
same time. In this second type of filter feeding only the head of the
larva was at the water surface and the rest of the body remained under
water.

In most browsing activities all or most of the mouthparts are
employed. When an object such as a long thin piece of decaying grass
comes into the feeding current, it comes in contact with the mouthparts
as follows:firstly, the serrated lateral labral brush hairs (median type 3)
hold a part of it, and push the remainder posteriorly; second, it slides
over the central labral brush; third, it passes between the epipharyngeal
bristles; fourth, the mandibular denticles strike it as it passes by, and
if a small piece of it is thus torn off it may go posteriorly with the current,
it may be drawn into the mouth, or it may settle on the prementum; fifth,
it passes between the maxillary brushes; sixth and finally, the particle
of grass touches the submentum and the aulaeum. During this process
some of the median labral brush hairs hold the particle while the remaining
hairs of the brush produce currents.

Sometimes parts of the lateral labral brushes move only slightly
(median type 3 hairs) whereas the hairs of their most posterior (types
2 and 3) move more actively. More commonly, all the hairs on the
brushes move simultaneously when producing a current. When a larva
comes to a stop after moving about in a container, it will gradually extend

surface view of currents

labral brush   maxillary brush

a

b

c
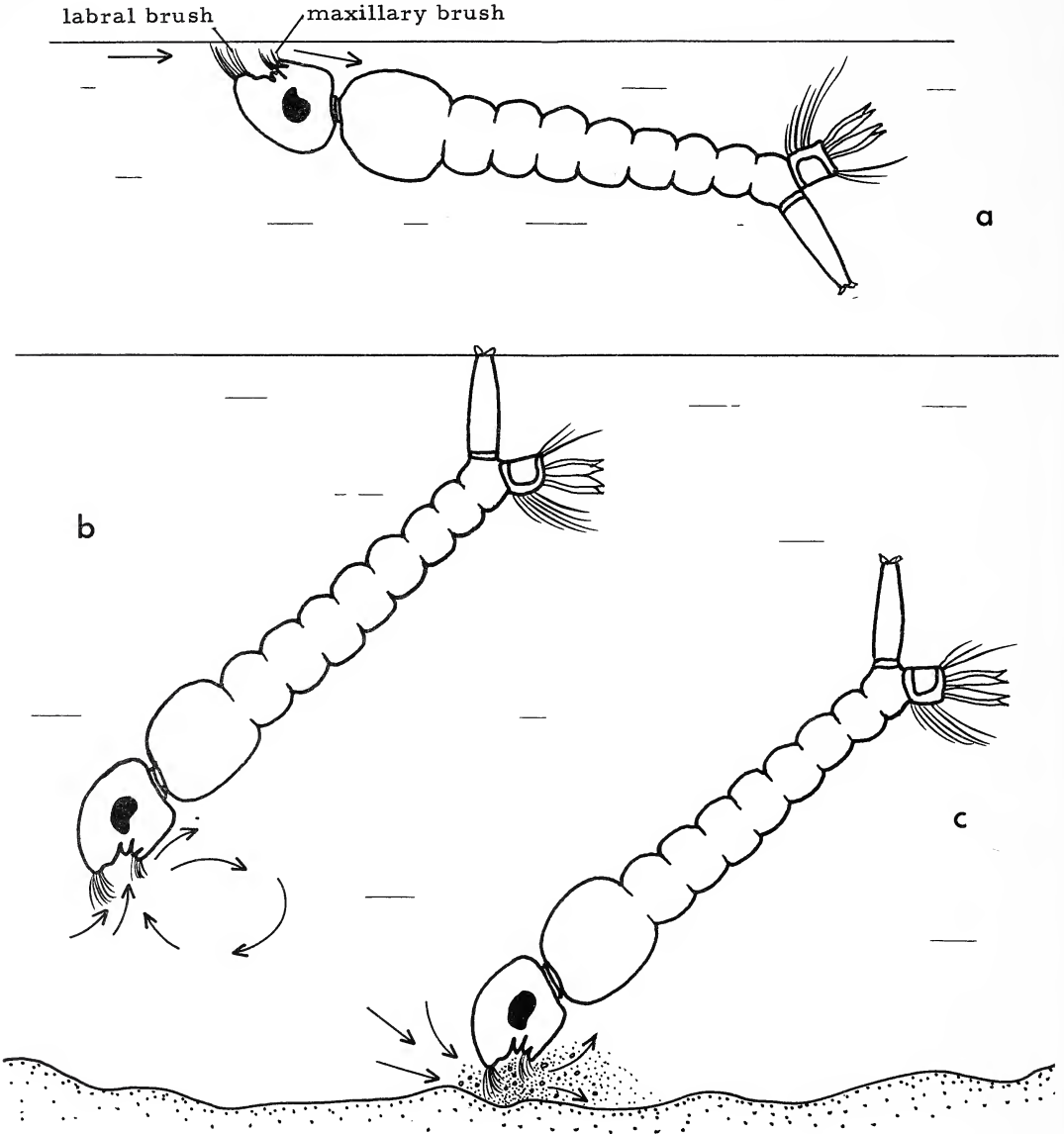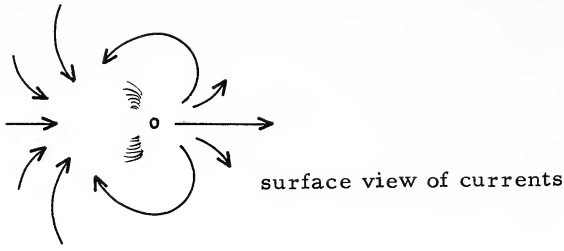
Fig. 8. Movements of labral brush currents of browsing larvae; (a) interfacial surface feeding current, (b) current produced under the water surface, (c) current used to stir up debris from the bottom.

or contract the brushes.

Most of the observations on the coordination of moving mouthparts were on *Aedes aegypti* and *A. fitchii* larvae which had been slowed down in a 20 to 30% solution of methocel of 400 centipoises. The larvae were watched in white porcelain spot plates with their ventral sides turned up. The following combinations of mouthparts were observed in action: 1. The lateral labral brushes moved in their usual antero-posterior oblique direction, and the long apical setae of both maxillae moved backwards and forwards at the same time. 2. The lateral brushes moved in their usual direction while the setae of one maxilla remained stationary, directed posteriorly, and the setae of the other maxilla continued their antero - posterior motion. The epipharyngeal bars also moved. 3. The lateral labral brushes were motionless. At the same time either one or both maxillary brushes waved and thus kept the current in motion. 4. The lateral labral brushes came to rest on the epipharynx and at the same time the other mouthparts moved in one of the following ways: one or both maxillae moved in the transverse plane; one or both mandibles moved in the transverse plane, striking against the hypopharynx; or, one mandible and one maxilla on the same or the opposite side moved. The same type of combination of mouthpart movements was observed in the larvae of the following species: *Aedes cataphylla, A. excrucians ,Aedes fitchii, A. hexodontus* Dyar, *A. punctor , A. riparius, A. sticticus , A. vexans* , and *Culiseta inornata.*

Aedes aegypti larvae also browsed on poplar and elm leaves in the laboratory. For two weeks ten larvae were given no other food but dried leaves of *Ulmus* sp. and no mortality occurred. At the end of the two week period all the larvae had pupated. The larvae of this species are also reared on leaves of a species of poplar in South Africa (Hocking, personal communication).

Browsing larvae of *Aedes* and *Culiseta* were observed in deep water pools (approximately 1.5 to 2.5 ft. deep) and in shallow pools (four to 12 in. deep). In shallow pools with clear water it was possible to see larvae browsing on submerged rotting leaves and other objects for as long as three minutes without coming to the surface for air. When the larvae came to the surface they sometimes remained there for one to five minutes and they either moved slowly or continued in one position before submerging again. Sometimes the wind disturbed the surface of the pool and some of the larvae that were at the surface moved with the wind, while others swam against this. In situations of this type, however, most larvae went to the edge of the pool, where a stable resting position was found.

Several observations of larval activity were made at a pool 1.5-2 ft. deep, and the courses of larval movements were recorded. *Aedes excrucians* and *Culiseta inornata* larvae were able to remain in a stationary position at the surface for from a few seconds to four minutes. During this time they probably produced currents with their mouthparts as did the larvae of these and other browsers when observed in a glass container in the laboratory. The approximate mean distance that any one larva covered in four minutes was between four and five feet. In a larger pool some larvae covered more space than this before submerging. The larvae went under either of their own accord, or wehn they came in contact with

another animal in the water such as a snail, a water beetle, a crustacean, or a dead insect floating on the surface. When larvae submerged without coming in contact with something first, after detaching their siphons from the surface film, they were pulled downward by the currents of their mouthparts.

In pools populated with browsing larvae and located in areas which were partly shaded, the shaded areas were much more crowded with resting larvae, although the sunny areas were used for moving about and browsing by a few larvae. This behaviour can be interpreted as orthokinetic (Fraenkel and Gunn 1960).

Filter Feeders

Three filter feeding species are at present known in Alberta, representing three genera; *Anopheles earlei* Vargas, *Culiseta morsitans*, and *Culex territans* . All of these species are uncommon, hence it was not possible to study the morphology and function of the mouthparts of their larvae in much detail.

Feeding larvae of *Anopheles earlei* were observed in the laboratory , but most were reared into adults and none was preserved for morphological study. *Anopheles earlei* larvae are small and are usually found in deep water, hence it is difficult to observe the action of their mouthparts in their natural habitat. In the laboratory all were usually at the water surface It was common to see some larvae resting with parts of the abdomen or thorax or both against the side of the container, while others moved in circular paths around the container. Often two or three larvae moved side by side in one direction, while one or more other larvae moved in an opposite direction. Sometimes two larvae, moving towards each other, collided and then both moved together in the direction initially travelled by one or the other. It is not known what determined the final direction of movement; perhaps the larva producing the stronger current overrode the other. *Anopheles* larvae turned their heads through 180 degrees so that the ventral side of the head was at the surface for period of 25 to 30 sec at a time as compared with approximately 10 sec periods in the normal position.

According to Clements (1963), The area of surface that can be cleared of particles by an *Anopheles* larva in a given time varies with the size of the larva, density of particles, and the rate of filtration, which is affected by temperature. The effect of these factors on larval movement was not considered in this study.

The movements and feeding behaviour of *Culex territans* and *Culiseta morsitans* are similar. The two species are found in the same type of habitat, and their mouthparts are similar in form. Since the labral brushes in these species are longer than in the *Aedes* or *Culiseta* browsers, the currents they create cover a larger area than do those of the browsers. Also these filter feeders extend their labral brushes mainly laterally, whereas the browsers extend theirs antero-laterally. Several *Culiseta morsitans* larvae were observed in a glass jar in the laboratory. They moved rapidly by means of the labral currents and fed at the same time; the pharyngeal movements could be seen through the head cuticle. Sometimes minute crustaceans were brought to the mouth with the current,

but they were not ingested. The food of the larvae consisted mainly of moss particles which floated in the pool water, and settled on the bottom of the jar. The particles on the bottom of the jar were agitated by browsing *Aedes cinereus* or *Culiseta inornata* larvae often collected with *C. morsitans*. Occasionally the *C. morsitans* moved their labral brushes just above the sedimented particles on the floor of the jar in the same manner as the browsing species. Sometimes two, three, or more of these filter feeding larvae rested in one location close to each other, clinging to the water surface film with their siphons, and moving their labral brushes. Most frequently the larvae stayed in such a position between two and three minutes before being disturbed by a moving larva or crustacean. When disturbed, the larvae either submerged, or moved horizontally on the water surface to another location. The first course was followed by about two thirds of the larvae. After submerging, each larva went in a different direction and stayed under the water surface for 10 to 15 sec. Upon coming to the surface the larvae either resumed their stationary positions for two to three minutes or until disturbed, or they moved horizontally, propelling themselves by the feeding current. In submerging when disturbed and in returning to the water surface the characteristic wriggling motion of the abdomen was used.

In the laboratory *C. morsitans* larvae assembled in the shady rather than the sunny part of a container. This observation is in agreement with that of Hocking (1953) on *Aedes communis*.

Predators

Three species of predatory larvae, *Chaoborus americanus*, *Mochlonyx velutinus*, *Eucorethra underwoodi* Underwood were collected near Flatbush, Alberta during the summers 1960 and 1961. *C. americanus* larvae were observed feeding on the larvae of several species of *Aedes* in the laboratory. The feeding behaviour of *Chaoborus* species has been studied in detail by Montchadsky (1945) and by Schremmer (1950). Both authors discussed the modification of the larval mouthparts for their predatory function. The mandibles in the larvae of this genus are the important movable mouthparts. The maxillae are fused with the ventral part of the cranium, and prementum is reduced to a wedge-like plate. The mandibles do not have a primarily crushing function, but their sharp strongly chitinized teeth have a holding and pushing function (Schremmer 1950). These larvae also use their prehensile antennae for catching prey. They ingest their prey whole. The main features of the mouthparts of *Chaoborus americanus* are indicated in table 3. The posterior occipital parts of the head capsule of *Chaoborus* larvae are connected to the subgena by membranes (Cook 1956); this permits the mouth opening to become enlarged whenever necessary.

In *Mochlonyx velutinus* larva the ventral part of the head is sclerotized, but a large mouth opening is present, as the head capsule is wider than in *Chaoborus*. Cannibalism was observed among the *M. velutinus* larvae in a jar in the laboratory. The raptorial function of the mandibles and antennae was observed when the larvae caught their prey tail first. Then the prey seemed to be held by the maxillae while the mandibles continued striking it and pushing it further into the mouth. In the specimens that I observed the process of ingestion lasted approximately two hours.

Digestion may take as long as three hours (Montchadsky 1945). Some-
times a feeding larva lost its prey, even if this was half ingested, if it
was disturbed by other organisms. James (1957) observed that *M. velutinus*
larvae are occasional predators on other mosquito larvae. I observed
M. velutinus feeding on larvae of various *Aedes* species. A similar habit
was observed in *M. culiciformis* De Geer by Montchadsky (1953) and
Montchadsky and Berzina (1959). Cannibalism was also observed in
Cryophila lapponica Mart. by Montchadsky (1953).

Discussion

The larvae that I studied in this investigation can be classified
as filter feeders, browsers, and predators. There are more similarities in
the structure and in the function of the various mouthparts of filter feeders
and browsers than between either one of these types and the predators.

The labral brushes of filter feeders and browsers are used for
bringing food to the larvae by means of currents which they produce by
vibrations. By means of these vibrations the larvae also move through the
water. The labral brushes of the predatory larvae are reduced to a few
bristles and do not produce currents.

The epipharynx of the browsing and filter feeding larvae is believed
to have the function of covering the mouth opening (Schremmer 1950).
This was not observed in the larvae that were studied in this project.
The epipharyngeal hairs were erected by the muscle which moves the
epipharyngeal bar, and when these hairs came in contact with the labral
brush hairs, food from the brush hairs was transferred to them. The
epipharyngeal hairs were in turn scraped by the mandibular hairs, and
this food was thus passed towards the mouth opening. If the food did not
go into the mouth, as often happened, particles of it remained on the pre-
mentum and on the hairs of the lacinia.

Mandibles of the browsing larvae were observed in actions of biting
while the larvae browsed. Those of predators were seen grasping and
pushing the captured prey into the mouth. The mandibles of the filter
feeders and the browsers move in a dorso-ventral plane, but those of
the predators move in an oblique plane which is nearly parallel to the long-
itudinal axis of the body.

LARVAL FOOD AND MOUTHPARTS

As a final step in investigating the function of the mouthparts the
nature of the food of the functional groups of larvae and the relationship
between the size of the food particles and the dimensions of the mouth-
parts were studied.

Procedures

The gut contents of several species of *Aedes*, *Culiseta*, and *Culex* lar-
vae were examined and measured. Most of these contents were dissected
out and mounted in glycerine jelly, a suitable preservative for plant mater-

ials (Sass 1940). Particles of activated charcoal were made available to several *A. fitchii* and *C. inornata* larvae, and ingested as well as uningested particles were measured.

The following measurements were taken of the larvae of available species,including *Anopheles*, *Chaoborus* , and *Mochlonyx*: head width (between the bases of the antennae), head length (between the median labral brush and the occiput), mean length of the right lateral labral brush (at the center of the brush), width of the right lateral labral brush (width at the base of the brush), and the width.of the epipharyngeal constriction (space between the most posterior, longest teeth on the transverse bars of the epipharynx).

An examination was also made of the material suspended in the water of a larval habitat. Ten litres of water was taken from a pool near Edmonton where *C. inornata* larvae were collected in September, 1961. This water was passed through a series of sieves. Material that did not go through the first sieve was examined, and a rough estimate of its composition was made. These fractions of material were then dried at 100°C to constant weight;they were ashed in a muffle oven at 575°C; the ash was weighed and the percentage loss was calculated.

Results

Table 6 contains a summary of the sizes of particles that were found in the guts and in the environment of the larvae of *Aedes fitchii*, *Culiseta inornata* and *Culex territans* . Particles that were identified from the guts of 4th instar larvae of these species are listed in table 7. From this table it is seen that the gut contents in the three species were similar.

The relationship between the structure of some mouthparts and the feeding habits of larvae is shown in fig. 9. The points on the graph were derived in the following manner:(1) for the position on the abscissa the mean length of the right labral brush was multiplied by its mean width to give the area swept by the brush. This product was divided by the product of the head lenght and the head width, to relate this to the size of the larva. (2) for the position on the ordinate the width of the epipharyngeal constriction was divided by the head width to represent the maximum relative size of particles which could be swallowed. Each point represents the mean value for a species. A separation between filter feeders and browsers is shown on this graph.

In fig. 9 the intermediates fall closer to the browsers than to the filter feeders. Typical filter feeders may be tentatively defined as larvae in which both the ratio of the epipharyngeal constriction to the head width and the relative area swept by the lateral labral brushes exceed 0. 14. In browsers and intermediates both of these ratios are less than 0.14. In typical predators the first ratio is more than 0.14, but the second is less. On the basis of morphology representatives of all types of feeders fall within the range of browsers.

From table 7 it is seen that the gut contents were similar in the three species, *Aedes fitchii* , *Culiseta inornata* , and *Culex territans* . The guts of a few *Chaoborus americanus* larvae that were examined were filled with muscle tissue; some of this was from other mosquito larvae.

TABLE 6 - Size ranges of particles in the guts and in the environments of 4th instar mosquito larvae. Percentage by number.

Max. linear dimension in microns	*Aedes fitchii*		*Culiseta inornata*			*Culex territans*	
	Charcoal in Water	Gut	Nat. food in gut	Natural food in Water	Gut	Charcoal in Gut	Nat. food in gut
< 7.5	4.3	6.1	3.1	2.7	4.0	4.0	12.0
- 9.9	7.2	10.2	7.4	9.8	5.7	8.2	35.4
- 14.9	29.5	31.7	6.3	6.0	10.1	27.1	40.1
- 19.9	11.1	10.0	27.2	35.6	30.3	9.1	7.2
- 24.9	9.0	8.0	11.5	9.0	12.0	9.5	6.3
- 29.9	13.0	9.8	9.3	10.0	11.7	12.2	
- 34.9	6.0	9.2	13.6	11.0	6.2	9.4	
- 39.9	4.9	5.6	10.1	8.5	4.0	3.9	
- 44.9	11.0	5.0	6.7	6.0	5.0	10.0	
- 71	5.0	4.0	5.3	2.0	10.0	8.0	
Nos of measurements	**500**	500	400	120	500	500	300

TABLE 7 - Organic particles in larval habitat and gut contents of 4th instar larvae of *Culiseta*, *Aedes*, and *Culex*; x scarce, xx common, xxx abundant, xxxx very abundant.

	Culiseta inornata		*Aedes fitchii*	*Culex territans*
	Habitat	Gut	Gut	Gut
Diatoms				
Fragilaria sp.	xx	xx	xx	
Gomphonema sp.	xx	xx	xx	
Navicula sp.	xx	xxxx	xx	
Pinnularia sp.		xx	xx	
Stauroneis sp.	xx	xx		
Green Algae				
Ankistrodesmus sp.	xx		xx	
Geminella sp.	xx	xx	xx	
Microspora sp.	xx		xxx	
Scenedesmus sp.	xx		xx	
Spirogyra sp.	xxxx	xx	xx	
Blue Green Algae				
Anabaena sp.			xx	
Gleocapsa sp.		xx		
Fungi				
Cladosporium spores	xx	xx	xx	
Rust - telospores			xx	xx

Fungi				
Rust - uredospores		xx		
Smut spores	xx	xx	xx	xx
Fungi Imperfecti, hyphae		xx		
Pollen of:				
Pinus				xx
Populus	xx	xx	xx	
Compositae	xx	xx	xx	xx
Plant Fibers				
xylem	xx	xx	xx	xx
tracheids	xx	xx	xx	xx
Flagellates				
Chlamydomonas sp.		x		
Euglena sp.	xx	xx alive		
Phacus sp.	xxx	xx alive		
Arthropod material				
Pieces of cuticle	xx	xx	xx	
Larval culicine				
spines, hairs	xx	xx	xx	

TABLE 8 - Particle size and weight in mg of suspended matter in 10 l of water from a pool occupied by *C. inornata* larvae.

Meshes/in					
	Passing		45	80	230
	Retained by	45	80	230	325
Dry weight (mg)		26.4	74.8	434.0	156.0
Ash weight (mg)		12.9	49.4	333.0	124.0
% organic matter		50	31	23	20

The following items were retained from the water taken from a pool were·*C. inornata* larvae were collected by a sieve with 45 meshes per inch: 60% *Cyclops* sp. and other copepods, alive; 20% decaying animal and plant material including mosquito eggs, egg cases beetle abdomens, and mosquito wings; 20% algae, mainly *Spirogyra* sp. The dry and ash weights and percentage of organic matter in the material held by sieves of finer mesh are given in table 8.

Discussion

In examining the gut contents of browsing, filter feeding, and predatory larvae it was found that the browsing *Aedes* and *Culiseta* larvae ingest items of similar types and sizes. The approximate proportions of

Fig. 9. The relation between feeding habits and morphology of mouthparts of mosquito larvae. ▲ predators, X intermediates, □ browsers, ○ filter feeders, Aedes, stipple Culex, black Culiseta, shaded Anopheles, 2 & 3, 2nd and 3rd instar C. inornata respectively.

the numbers of food particles of the different sizes in the guts of *Aedes fitchii* and *Culiseta inornata* larvae are: less than 15μ, one - sixth , 15 - 22μ, one-third, 22-40μ, one-third, 40-71μ, one-sixth of the measured particles.

In the larvae of *A. fitchii* 58% **of** the charcoal particles ingested were found to be less than 20 microns with the largest percentage (31.7) in the 10-15μ range;only 6.3% of the natural food particles fell in the 10-15 μ range with the largest percentage (27.2) occurring in the 15-20μ range. A similar relationship was found in *C. inornata* (table 6).

Some plant and animal particles were folded before entering the mouth of the larva. Also when the larvae browsed on plant surfaces they bit pieces off plants, scraped surfaces, and thus obtained soft particles of various sizes and shapes. Many plant particles eaten were long, narrow, and flat, so they were easily carried into the mouth by the feeding current. However, when activated charcoal was placed in the water, the larvae ingested the small particles that were brought to the mouth with the feeding current, but did not take in the large ones which rapidly settled on the bottom of the container. Charcoal particles are denser than natural food and the browsers' currents cannot stir up particles larger than 15 microns. The particles are filtered by the labral brushes; large hard particles are rejected, whereas soft food is actively taken in. Occasionally I stirred the charcoal in the containers. Sometimes the larvae browsed on the bottom of the container, but long, flat particles were difficult to obtain. Thus mostly small charcoal particles were scraped into the mouths.

Since the charcoal particles did not remain in water suspension very long, they were not fed to the filter feeders. Pond food from the guts of these larvae was measured (table 6). Also measured were the spaces between the groups of labral brush hairs through which the feeding current passes. The size of these spaces was found to be similar to that of the particles in the guts. Thus filter feeding is possible among these larvae, for if the ingested particles were larger than the spaces between the hairs, they would not be trapped in the brushes, but would remain on the surface of the brush. On the other hand, very small particles would pass through the brush with the water current without becoming entangled in it.

Also, most of the food particles found in the guts of filter feeders were of the same order of size as the charcoal particles ingested by the browsers, and smaller than the food particles of browsers that fed in the field.   The epipharyngeal constriction width in filter feeders is greater than in browsers, therefore it should permit larger particles to pass to-wards the mouth. However, the mandibular teeth of filter feeders are weakly sclerotized and cannot crush or "squeeze" large particles in the feeding current. Thus large soft particles by–pass the mouth openings of filter feeders, whereas they are pushed into the mouths by the mandibular teeth of browsers. But the wide epipharyngeal space of filter feeders allows the passage of more particles in a given time.

According to Bates (1949), Shipitzina in 1935 found that 4th instar larvae of *Anopheles messeae* Fall. were able to swallow sand particles from 68-165μ wide.   The mouth openings of this species must be larger than **those of the culicine larvae** I studied.   The size range of food particles found in the guts of three English species of *Simulium* larvae was found

to be 1. 7-15. 1µ by Williams *et al* (1961), the size of the mouth openings of these larvae was not given.

McGregor (1963), working with larvae of *Opifex fuscus* found that first instar larvae did not develop serrations on their labral brushes if they were fed on minute particles of dehydrated blood serum. Serrations did develop when they were given fish food ranging in particle size 0. 1-0. 6 mm. Similar experiments with larvae of other feeding types should be revealing.

The browsing larvae whose guts I examined fed on plant particles and on microscopic animals, whereas the filter feeder *Culex territans* had fed only on plant particles (table 7). Also, all the types of particles that were present in the pool water where the *C. inornata* larvae were collected were found in the intestines of these larvae. It can be said then that these larvae do not discriminate in the type of food they ingest. Other workers have come to similar conclusions: Coggeshall in 1926 as reported by Bates (1949), Howland (1930), and Jones (1960) who worked with anopheline larvae, and Becker (1958) who worked with larvae of *Culicoides circumscriptus* Kieff. These authors have found algae, diatoms, and other plant particles in the guts of *Anopheles* and *Culicoides* larvae. Rempel (1936) found similar food materials in larvae of *Chironomus hyperboreus* Staeg. (= *C. rempelii* Thienemann, Rempel 1962). Other culicine larvae also ingested similar food (Horsfall 1955). Bekker (1938b) found living *Euglena* in the gut of *Anopheles maculipennis*

The *Aedes* and *Culiseta* browsers show similarities in both function and morphology. The range of the ratio of epipharyngeal constriction to head width is from 9 to 12. 7, and the ratio of the area swept by the lateral labral brushes to the head size ranges from 4 to 11. 8 (fig. 9). Two *Anopheles* filter feeders, one *Aedes* intermediate, and two *Culex* intermediates also fall within these ranges. The second ratio is even higher for another intermediate feeder; it is 13 for *Aedes cinereus*

Of the species I examined, two species of *Culex* and one of *Culiseta* are filter feeders in function and morphology. The species of *Chaoborus* and *Mochlonyx* are predators both functionally and morphologically. The remainder of the species represented in fig. 9 range between these two types either in function, morphology, or both. Thus the *Aedes* and *Culex* species labelled as intermediates obtain their food by filtering, but the structure of their mouthparts is intermediate between the typical filter feeders and typical browsers. The *Anopheles* species are also filter feeders. Their mouthparts fit the general description for filter feeders but the sizes of the mouthparts measured, upon which the division in fig. 9 is based, are proportionately smaller than the sizes of corresponding mouthparts of *Culex* and *Culiseta* filter feeders.

While this method of separating larvae of *Aedes* , *Culex* , and *Culiseta* , into filter feeders and browsers is satisfactory and can be used to categorize the predatory species of *Chaoborus* and *Mochlonyx*; it is not reliable for *Anopheles* . The filter feeding larvae are considered to be the most primitive and the predatory larvae the most advanced (Montchadsky 1937, Surtees 1959). Thus the largest number of the species studied are in a transitional stage of evolution.

GENERAL CONCLUSIONS AND DISCUSSION

According to the functions of the mouthparts three types of mosquito larvae can be recognized: filter feeders, represented in Alberta by *Anopheles earlei* , *Culex territans* , and *Culiseta morsitans*; browsers, including most of the *Aedes* and *Culiseta* species; and predatory, represented by species of *Chaoborus*, *Mochlonyx* , and *Eucorethra*. The *Culex* and *Culiseta* filter feeders are characterized by labral brushes consisting of long, thin, simple hairs, and by lightly sclerotized mandibles. The *Anopheles* larvae have thin, simple lateral labral hairs which are shorter than those of *Culex* and *Culiseta*, slightly sclerotized mandibles, and large rectangular maxillae with short thin hairs. The browsers have shorter labral brushes with some serrated, thick hairs, rectangular maxillae with shorter, thicker brushes, and moderately sclerotized mandibles. The predators bear only a few setae on their reduced labral areas and on their much more fused maxillae, and they have heavily sclerotized mandibles.

Among the browsers morphological intermediates occur. *Aedes canadensis* and *A. cinereus*, have short labral brushes with simple hairs , browser-like mandibles, and maxillae similar to those of the filter feeders. *Culiseta impatiens* and *Culiseta inornata*, have typical browsing labral brushes and mandibles, but have maxillary structures closely related to those of predators.

Not much variation was observed in the structures of the labral brushes, mandibles, or maxillae among most of the browsing *Aedes* larvae studied. However, specific differences were found in the numbers of serrations on the sclerotized plates of the prementum, and on the triangular submentum. These characters may be taxonomically useful.

By staining with Mallory's triple stain it was found that the cuticle of the mouthparts varies in hardness and flexibility. The median hairs of the lateral labral brushes of the browsers have hard basal and central parts, and flexible parts just above the bases, and at the tips.

An examination of larval food revealed that the browsing and filter feeding larvae are not discriminatory in the type of food they accept, but there are limits in the size of particles they can ingest.

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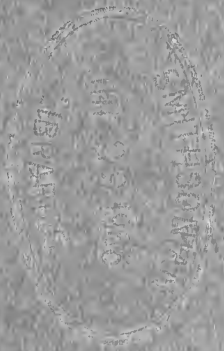
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A REVISION OF THE NORTH AMERICAN SPECIES OF THE *CICINDELA MARITIMA* GROUP WITH A STUDY OF HYBRIDIZATION BETWEEN *CICINDELA DUODECIMGUTTATA* AND *OREGONA*

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The North American species of the Cicindela maritima group are: C. duodecimguttata *Dejean*; C. oregona *LeConte*; C. depressula *Casey*; C. repanda *Dejean*; C. hirticollis *Say*; C. limbata *Say*; C. columbica *Hatch*; C. bellissima *Leng*; and C. theatina *Rotger. The male genitalia of these species are described. The group is diagnosed and two keys are given, one based on non-genitalic characters, and the other based on features of the male genitalia. For each of the species* duodecimguttata, oregona, *and* depressula *the following are presented: synonymy, analysis of geographic variation in size, coloration, color pattern of elytra, and distribution. Descriptions of the subspecies of* oregona *and* depressula *are given. Hybridization between the species* duodecimguttata *and* oregona *is examined quantitatively by means of the hybrid index method and the data are presented in the form of histograms. The zone of hybridization lies on the eastern slopes of the Rocky Mountain System from Colorado to the Northwest Territories, and is about 50 miles wide in Alberta but is nearly 1,000 miles wide in northern Canada. Variation of external characters and shape of the median lobe of the male is greater in the region of intergradation than it is within the range of the pure parental forms. Temporal variation occurs in hybrid populations. Phylogenetic and zoogeographic relationships are postulated to explain the structural similarities and distribution patterns of the North American species of the* maritima *group.*

INTRODUCTION

The species of the North American tiger beetles of the genus *Cicindela* are for the most part fairly well understood taxonomically, and it is possible to identify most adult specimens as a result of publications by Leng (1902), Horn (1915), Cazier (1936, 1948, 1954, 1960), and Wallis (1961). In addition Hamilton (1925) has described many larvae. With the descriptive phase in this condition attention must now be directed to taxonomic studies at the species level. By such studies phylogenetic relationships of species and delimitations of species groups within the genus can be worked out.

This study began with the discovery of hybridization between *Cicindela duodecimguttata* and *Cicindela oregona*. Variation of phenotypic characters of hybrids and pure parental forms was analysed. As a result it was found that the latest definition of *oregona* (Wallis, 1961) was composite and included the definition of *depressula*. This led to a study of *depressula*. The male genitalia of all the North American species of the *maritima* group were then examined. The features of the internal sac proved to be diagnostic of this group, while shapes of the median lobes were found to be specifically distinct.

MATERIALS AND METHODS

Materials

Structural features and their variation were studied in approximately 12, 000 specimens of *C. duodecimguttata, C. oregona* and *C. depressula.* The data from these observations are analysed quantitatively by the following techniques. Descriptions and specimens of larvae of seven species were also examined but only as additional material for deriving a phylogenetic scheme for the North American species of the *maritima* group.

Methods

External Characters

Distribution of hairs on the head, elytral pattern, and color are very important in the classification of the North American species in the *maritima* group.

Hairs may cover the head and frons either very densely or sparsely, or they occur in the form of a patch near the inner edge of each eye. The post genae may be glabrous or hairy.

Elytral pattern is composed of the following white markings: humeral lunule, marginal band, middle band, and apical lunule. The occurrence, shape, and expanse of these markings are used in showing interspecific and intraspecific variation (figs 11-16).

The six basic colors that occur in *duodecimguttata, oregona* and *depressula* were matched with the color standards of Ridgway (1912). They are listed below with their corresponding Ridgway names in parentheses: black (Black), brown (Mummy Brown), copper (Liver Brown), purple (Dull Violet Black), blue (Dusky Green Blue), and green (Danube Green). These colors may be dull, opalescent, or metallic.

Male Genitalia

North American species of the *C. maritima* group can be grouped together and individually identified by characteristics of the male genitalia. For study of the genitalia the male beetle was relaxed in boiling water. Then by inserting a pair of fine forceps into the end of the abdomen the genitalia were grasped and pulled out. These structures were cleared in a hot 10% solution of potassium hydroxide for about 10 minutes and then washed in water. The genital structures were finally stored in glycerine in a corked microvial and pinned beneath the beetle from which they were extracted. Drawings of the male armature were made with the aid of a Wild camera lucida and stereoscopic microscope at a magnification of X 62.5.

Measurements

Intraspecific variation of size and expanse of color pattern was

analysed by means of measurements. A calibrated eyepiece in a Zeiss stereoscopic microscope at a magnification of X 10 was used. Relative size is indicated by length of elytra as measured from the apex of the scutellum to the tip of the elytral spine and width of the elytra as measured from the midline to the margin at the widest point. Expanse of elytral pattern is represented by the transverse diameter of the apical dot. Measurements were made of specimens in each large population sample.

Statistical Methods

Linear measurements were treated statistically and tabulated. The range, mean, standard deviation, coefficient of variation, and standard error, were determined in each analysis. The Chi-square test was used in evaluating data of annual and seasonal changes in hybrid indices of population samples. The method was also employed to assess randomness of mating in the hybrid zone (Simpson *et al.* 1960, p. 306).

Hybrid Index

Variation in a hybrid population can be analysed using a hybrid index. This method was developed by Anderson (1949) for plant hybrids and has been successfully used for study of variation in avian hybrid populations (Sibley 1950, 1954, Sibley & Short 1959a, 1959b, 1964, Short 1963). The hybrid index method makes it possible to describe variation in quantitative terms. The hybrid index is constructed as follows. Characters that separate the parental forms are determined. Each character of one parent is scored 0. Those of the other pure parent are each given a high value and intermediate characters are ascribed values that fall on the scale between the parental scores. The hybrid index for each specimen is the sum of its individual character values.

The method was used to analyse variation in *duodecimguttata - oregona* hybrid population samples. Results are presented in figures 35 to 44. In addition, geographic variation in elytral pattern of *duodecimguttata* is analysed by this technique (table 3). Because of its broader application the hybrid index is here also referred to as the "compound character index". In figs 35 to 43 average index changes per mile are indicated between localities of population samples. These roughly illustrate relative spatial rates of index change, but they do not imply linear trends.

Pictorialized Scatter Diagrams

Pictorialized scatter diagrams, the alignment of symbols in a two-dimensional field or graph, are used to describe several character relationships. The positions of symbols are determined by the calibrated axes each of which is a quantitative expression of a single character or ratio of two characters. More characters can be considered at a time by adding appropriate tokens to the specimen symbol. This method is used to illustrate data on intraspecific relationships of *oregona* populations (figs 22 - 31). For a more complete description of this method see Anderson (1949).

Pie-graph Maps

This method illustrates geographic relationships of populations with different varying color characters. Pies plotted on a map represent geographic positions from which population samples were collected. Numbers of specimens of particular color combinations are indicated opposite the appropriate pie sections (figs 19, 20).

Field Methods

Because tiger beetles are rather difficult to see in their natural environment a technique was necessary to facilitate field observations. At Nordegg, Alberta, in the hybrid zone, specimens were first caught with an insect net. The sex and hybrid index value of each individual was translated into a code that was painted on the elytra with a small brush. The individuals were then released and observed through field glasses.

Adult tiger beetles, collected for museum material, were killed in a jar containing potassium cyanide, and pinned the day they were caught.

Larvae were either trapped at the tops of their burrows by rapidly driving a shovel beneath them, or dug out. They were boiled in water five minutes to preserve their color and then placed in 70% alcohol.

Criteria for Species and Subspecies

Two similar forms are regarded as distinct species if their geographical ranges overlap and if they show no intergradation in at least one character (color excluded). If a narrow stabilized hybrid belt is developed in the region of contact of two forms that are largely allopatric they are treated as distinct species (Mayr 1963). Two allopatric forms that differ only in coloration are judged to be conspecific. Allopatric forms of a single species are regarded as being subspecifically distinct if 75 per cent of the individuals of one form are different from 97 per cent of the individuals of the other (Mayr *et al* 1953). However, if a clinal series of intermediate populations is intercalated between two distinct populations that are widely allopatric subspecific names are not given.

There are two opposing views regarding the subspecies concept. Wilson and Brown (1953) believe the subspecies concept to be subjective and arbitrary in the light of discordant variation, variation in microgeographic races, and the artificiality of quantitative methods of defining the formal lower limits of the subspecies as well as other reasons. Inger (1961) however, argues that Wilson and Brown tend to magnify exceptional cases, and that the subspecies concept despite its limitations has proved useful. It is this latter view that is followed in this text. Many more opinions regarding the subspecies concept are expressed in issues of Systematic Zoology (1953-1960).

MORPHOLOGY OF THE MALE GENITALIA OF THE NORTH AMERICAN SPECIES OF THE *CICINDELA MARITIMA* GROUP

Introduction

Several papers dealing wholly or in part with the male genitalia of American tiger beetles have been published (Horn 1930, Papp 1952, Rivalier 1954, and Rumpp 1957). Horn observes that for some races of *Omus californicus* Eschscholtz shape and size of the penis is characteristic. Papp presents a detailed study of the internal sac from which relationships of the Nearctic and Palearctic tiger beetles are deduced, while Rivalier classifies the entire *Cicindela* fauna of the Americas. Rumpp uses male genitalia in separating more clearly the species *Cicindela praetextata* LeConte and *Cicindela californica* Menetries.

Male genitalia of three or more specimens of each North American species of the *maritima* group were examined. The male armature consists of three relatively large sclerites: a median structure called the median lobe, penis, or aedoeagus (see fig. 1); and a pair of lateral parameres, one on each side of the median lobe and articulating with its base. Inverted in the median lobe is the membranous internal sac that is everted from the dorso-apical portion of the median lobe during copulation.

Within each species the shape of the median lobe is quite uniform. There is, however, a considerable amount of interspecific variation, particularly in form of the apex, that proves useful in distinguishing species of the *maritima* group from one another.

The internal sac comprises many folds, dark areas bearing microtrichia or aculeae, and sclerites. These fields of aculeae, and sclerites can be homologized within the species of the *maritima* group. Numbers are assigned to sclerites and letters are assigned to fields. This system of nomenclature follows that of Ball (MS) and is not synonymous with that of Papp.

When retracted in the abdomen the median lobe lies lengthwise, parallel to the longitudinal plane of the body of the beetle, and the opening of the internal sac is dorsal. When the median lobe is in a copulatory position the aperture of the internal sac is ventral. For each species drawings of the retracted median lobe and the inverted internal sac viewed from the dorsal and left sides are presented. In addition the shape of the apex of the median lobe is given separately for each species. Included also is a table of the various sclerites of the internal sac for each North American member of the *maritima* group.

Descriptions

Male Genitalia of Cicindela duodecimguttata Dejean

The median lobe is of average breadth and length (figs 1a, b, c, and 10). Two broad, lateral flanges that occupy the apical region of the median lobe converge apically to form a marked tip which curves

ventrally.

The inverted internal sac, in which three fields of aculeae are distinct is clearly visible. These darkened areas are labelled a, b, and c. Field *a*, which has a pebbly appearance is apical in the infolded position but it is basal when the internal sac is everted. Field *b*, ventral in position, is a finger-like projection of the membrane from which only the apical end is separate in the form of a tiny sac. When the internal sac is inverted *b* hangs inward whith its free end nearest the apex of the median lobe. Conversely, when the internal sac is everted, this field projects outward its blind end remaining oriented toward the apex of the median lobe. Field *c*, is three quarters circular, anterior in the infolded position, and appears to serve as the apical limits of the everted internal sac.

Six sclerotized areas are present. Most noticeable is the flagellum (4) which is a slender strip, pointed apically, and widened and hooked at the base. A short rectangular sclerite (3) is present to the left of the base of the flagellum, and sclerite 5, a cuplike structure, lies posterior to 3. In fig. 1 sclerite 5 lies to the right of the median line beneath several membranous folds, but it is more clearly shown in fig. 2. To the left of and lying in part over the basal portion of the flagellum is sclerite 1, a quadrate plate. Sclerite 6 is oriented to the right of the sagittal plane. It is twisted basally and resembles an aculeate field apically. Sclerite 2 is an elongate curved strip with its apical end near that of the flagellum. A very small triangular sclerite is present between 2 and 6, but it is not numbered since it may be a disconnected piece of one of these two sclerites. This sclerite is illustrated with sclerite 2 in fig. 10.

Male Genitalia of Cicindela oregona LeConte

Shape of the median lobe (figs 2a, b, c, and 10) is quite different from that of the preceding species. Though the apical, lateral flanges are about as long as those of *duodecimguttata* they are rather narrow. The apex is not markedly curved ventrally. Fields and sclerites, excepting sclerite 5 which is relatively large, look like their counterparts of *duodecimguttata*.

Male Genitalia of Cicindela depressula Casey

Unlike the penes of *duodecimguttata* and *oregona* the apical portion of the median lobe in this species is characterized by wide lateral flanges that form a blunt tip (figs 3a, b, c, and 10). The flanges are continuous and not separated from one another by the raised apical section of the chamber containing the internal sac as they are in the two preceding species. The median lobe is short and broad. Field *a* is composed of several elongate folds that together form a rough area. Field *b* is comparatively light, and *c* is c-shaped. Sclerites 1, 2, 3, 4, and 6 are respectively of the shapes and in the positions described for those of *duodecimguttata*. At the basal end of sclerite 2 the small triangular sclerite is elongate. Sclerite 5, large and lightly sclerotized, is visible when the internal sac is everted or inverted.

Fig. 1. Median lobe and inverted internal sac of *Cicindela duodecimguttata.* 1a, dorsal aspect; 1b, left lateral aspect; 1c, apical portion, dorsal aspect. Numbers = sclerite nos. Lower case letters = fields.
Fig. 2. Median lobe and inverted internal sac of *Cicindela oregona.* 2a, dorsal aspect; 2b, left lateral aspect; 2c, apical portion, dorsal aspect.

Male Genitalia of Cicindela repanda Dejean

      The portion of the median lobe that contains the internal sac is more apically confined than those of the three discussed species (figs 4a, b, c, and 10). The lateral flanges are narrow and widely separated dorsally by the chamber of the internal sac. Field *a* is small and lightly aculeate, while *b* is a distinct area pebbly in appearance. Field *c* is of the common shape. Sclerite 1 is large and triangular, while 2, 3, 4, and 6 are like those of *duodecimguttata*. There is no small sclerite near the basal end of 2. Sclerite 5, large and heavily sclerotized, is quite distinct.

Male Genitalia of Cicindela limbata Say

      The median lobe is relatively short and narrow (figs 5a, b, c, and 10). The two broad, lateral flanges are evenly rounded and together converge to a marked but non-protruding apex. Fields *a* and *b* are strongly aculeate; and *c* is clearly indicated in the form of one third of a circle. Variation is evident in the shape of sclerite 1 which is generally smaller in size than those of the other North American species of the *maritima* group. Sclerites 2, 3, 4, and 6 are of the common shape, and sclerite 5 is absent.

Genitalia of Cicindela bellissima Leng

      The median lobe is of average length but thicker than those of the preceding species (figs 6a, b, c, and 10). From a dorsal view the lateral flanges compose a broad apical region that terminates as a sharp projecting tip. Field *a* is clearly indicated by its dark compact appearance. Both *a* and *b* have large and scale-like aculeae. Field *c* is three quarters of a circle. Sclerites 1, 2, 3, 4, and 6 are each of the common shade intensity and shape. Sclerite 5 is absent.

Male Genitalia of Cicindela columbica Hatch

      The median lobe is relatively long and slender (figs 7a, b, c, and 10), the apical region comprises two fairly wide lateral flanges that are slightly constricted basally, and an unprojected, rounded apex. Prominent aculeae occur on field *a*, which is smaller and less compact than that of *bellissima*. Field *b* is a lightly shaded area, while *c* is of the common type. Sclerites 1, 2, 3, 4, and 6 each resemble their counterparts in other species of the *maritima* group. The sclerite between 2 and 6 is large and heavily sclerotized. Sclerite 5 is small and field-like in appearance which makes it difficult to detect.

Male Genitalia of Cicindela hirticollis Say

      The median lobe is elongate and thick (figs 8a, b, c, and 10). The chamber which contains the internal sac is extended dorso-apically so that the lateral flanges are widely separated, and the apical portion of the median lobe is obscured when viewed from the dorsal side. Field *a* is composed of several elongate folds; *b* is sparsely aculeate;
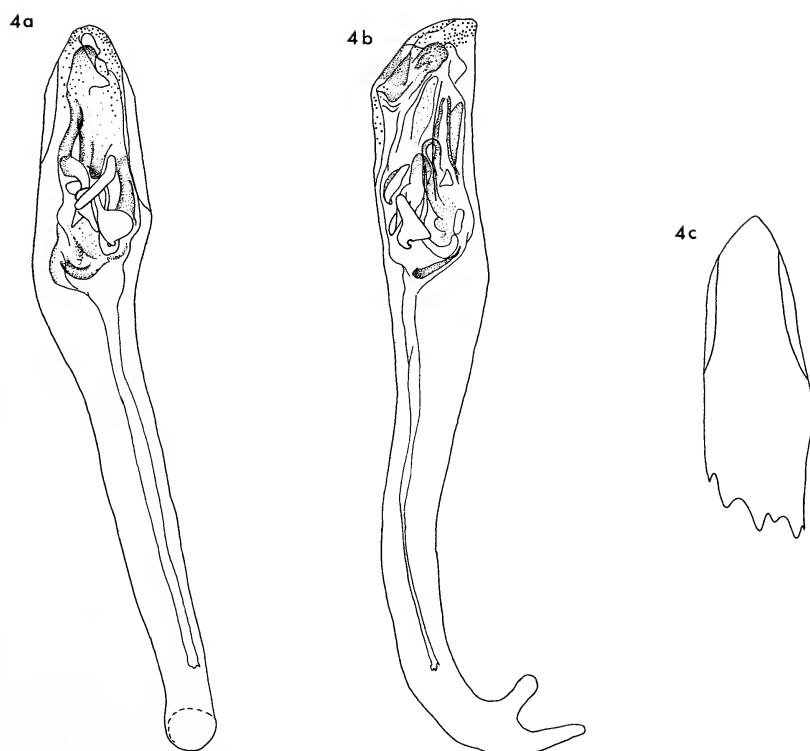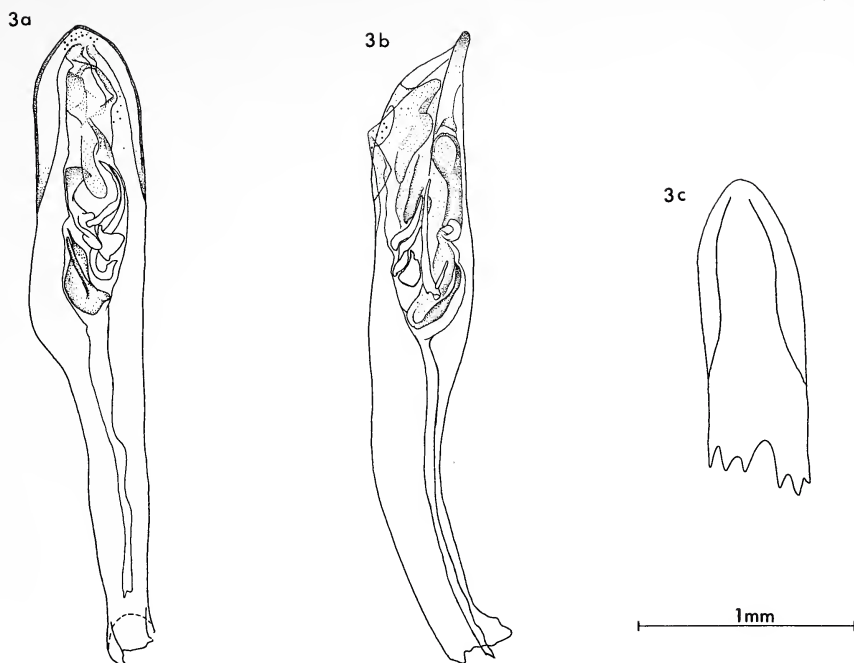
Fig. 3. Median lobe and inverted internal sac of *Cicindela depressula*. 3a, dorsal aspect; 3b, left lateral aspect; 3c, apical portion, dorsal aspect.
Fig. 4. Median lobe and inverted internal sac of *Cicindela repanda*. 4a, dorsal aspect; 4b, left lateral aspect; 4c, apical portion, dorsal aspect.
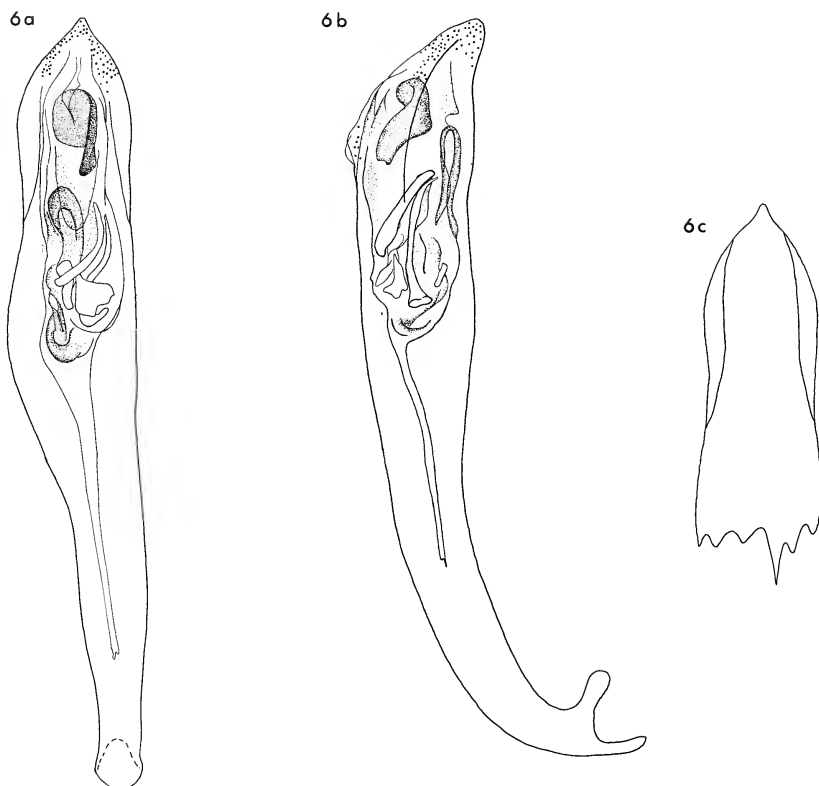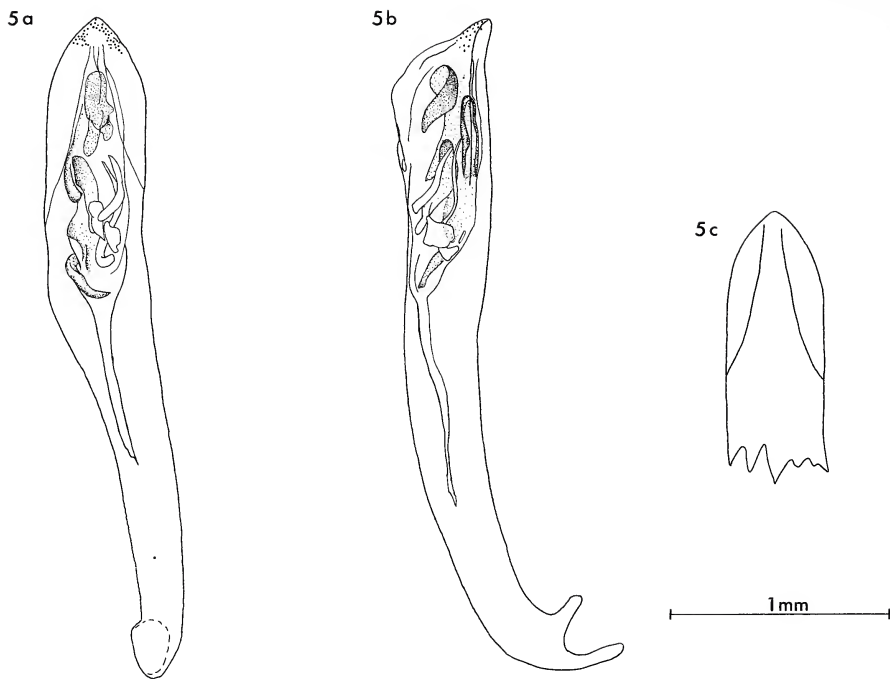
Fig. 5. Median lobe and inverted internal sac of *Cicindela limbata.* 5a, dorsal aspect; 5b, left lateral aspect; 5c, apical portion, dorsal aspect.

Fig. 6. Median lobe and inverted internal sac of *Cicindela bellissima.* 6a, dorsal aspect; 6b, left lateral aspect; 6c, apical portion, dorsal aspect.

and *c* is a semicircle. Sclerite 1 is relatively lightly sclerotized. Sclerites 2, 3, and 6 each have the shape characteristic of the *maritima* group. The sclerite between 2 and 6 is large and rectangular much like that of *columbica*. The apical twirl in sclerite 4 is markedly pronounced. Sclerite 5 is broad and lightly sclerotized.

Male Genitalia of Cicindela theatina Rotger

The median lobe is of average length and breadth (figs 9a, b, c, and 10), the apical region somewhat resembles that of *duodecimguttata* without the protruding tip. A distinct keel is present on the ventral apical portion of the median lobe. Fields *a* and *b* are strongly microtricheate and *c* is normal. Sclerites 1, 2, 3, 4, and 6 are of the general shape and size. The sclerite between 2 and 6 is large. Sclerite 5 is barely visible and only occurs as a small roughened area.

Discussion

It is difficult to fix the genitalia of each species in the same relative position for drawing purposes. Thus sclerites that are of the same shape but drawn in different positions may appear to be different from one another. The shapes of sclerites 2, 3, 4 (excepting that of *hirticollis*), and 6 are remarkably constant throughout the North American *maritima* group. This uniformity in sclerite shape sets these species apart as a unit from other *Cicindela* groups. Some interspecific differences of the internal sac are evident, however. These are: shape and size of sclerite 1; presence and size, or absence of the sclerite between 2 and 6; presence and condition, or absence, of sclerite 5. The shape of the median lobe is diagnostic for each species, particularly the form of the apical region viewed from the dorsal or ventral sides.

Median lobes and internal sacs of specimens taken in the hybrid region of *duodecimguttata* and *oregona* were examined. It was found that the form of the apex of the median lobe changed through intermediate shapes from pure *duodecimguttata* to pure *oregona*.

TAXONOMY OF THE NORTH AMERICAN COMPONENTS OF THE *CICINDELA MARITIMA* GROUP

Diagnosis of the Group

At the present time there is no generally accepted definition of the *maritima* group (*repanda* group, in part). Leng (1902) defined the *repanda* group on the basis of external characters in which *repanda, hirticollis, oregona,* and *duodecimguttata*, were brought together, but *limbata* and *bellissima* were excluded. Casey (1913) formed the *repanda* group on the basis of body size and shape of humeral lunule. The species *limbata* and *bellissima* were not included, and *hirticollis* was regarded as constituting a closely related but separate group. Papp (1952) used characters of the internal sac of
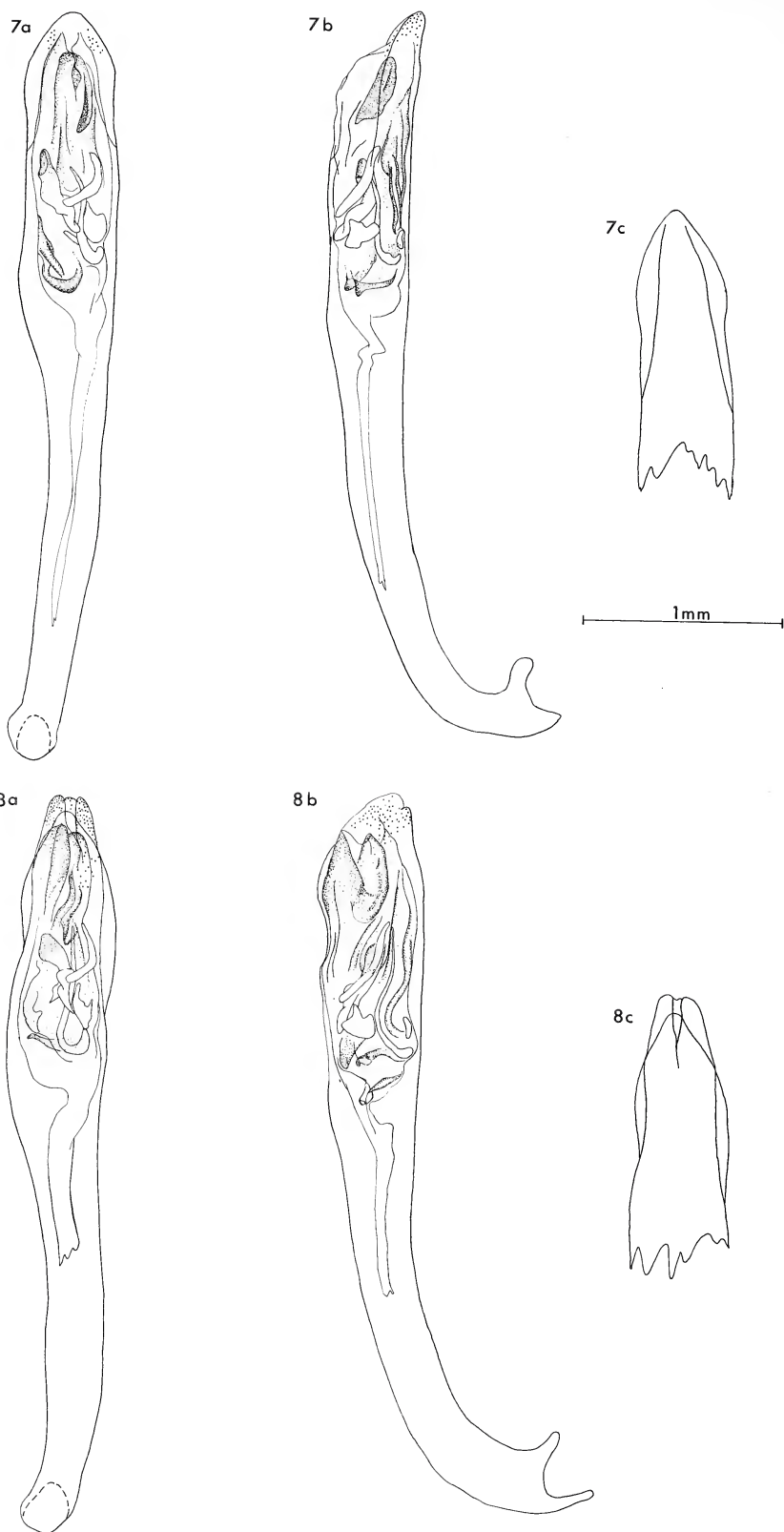
Fig. 7. Median lobe and inverted internal sac of *Cicindela columbica*. 7a, dorsal aspect; 7b, left lateral aspect; 7c, apical portion, dorsal aspect.

Fig. 8. Median lobe and inverted internal sac of *Cicindela hirticollis*. 8a, dorsal aspect; 8b, left lateral aspect; 8c, apical portion, dorsal aspect.
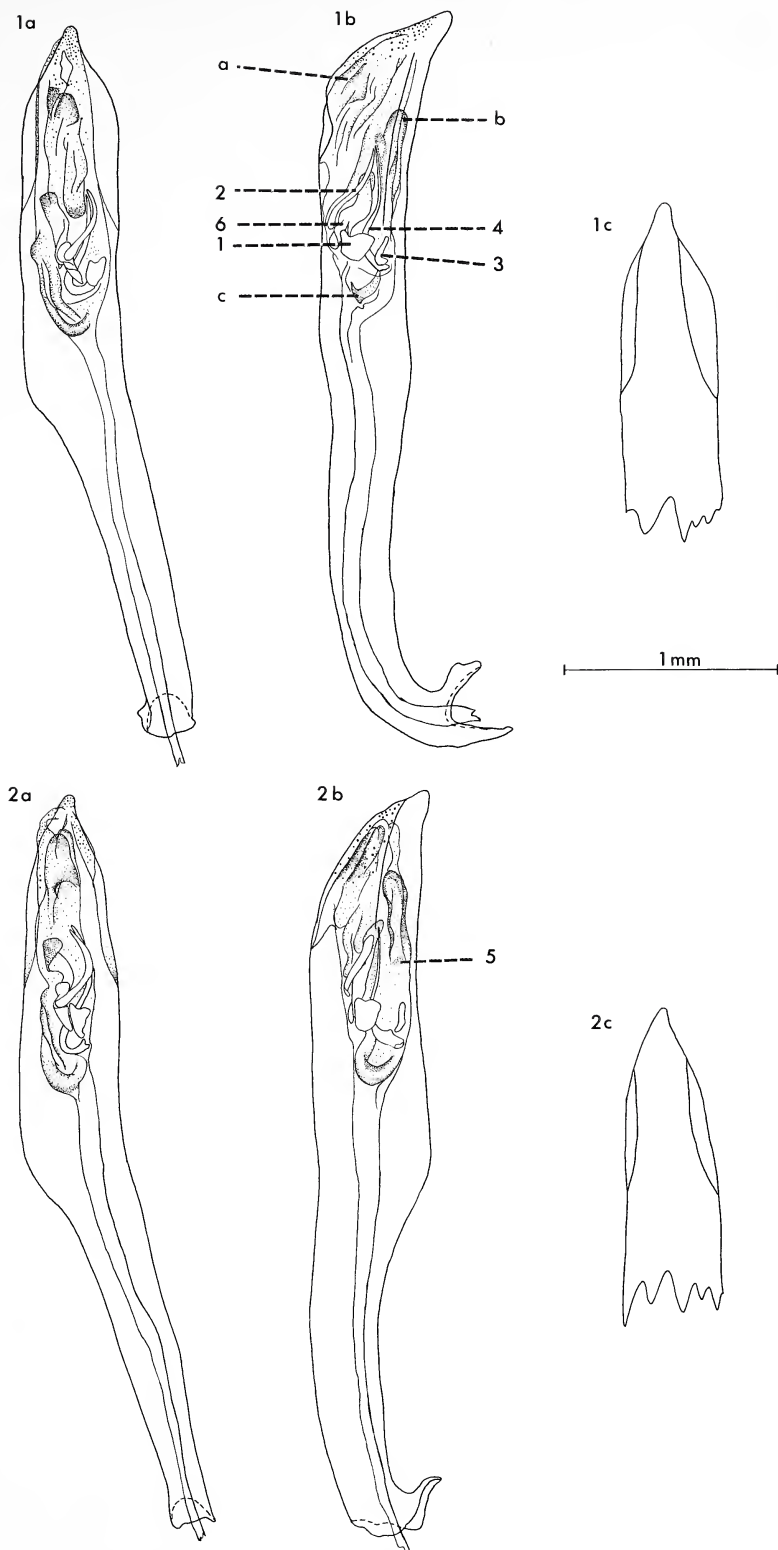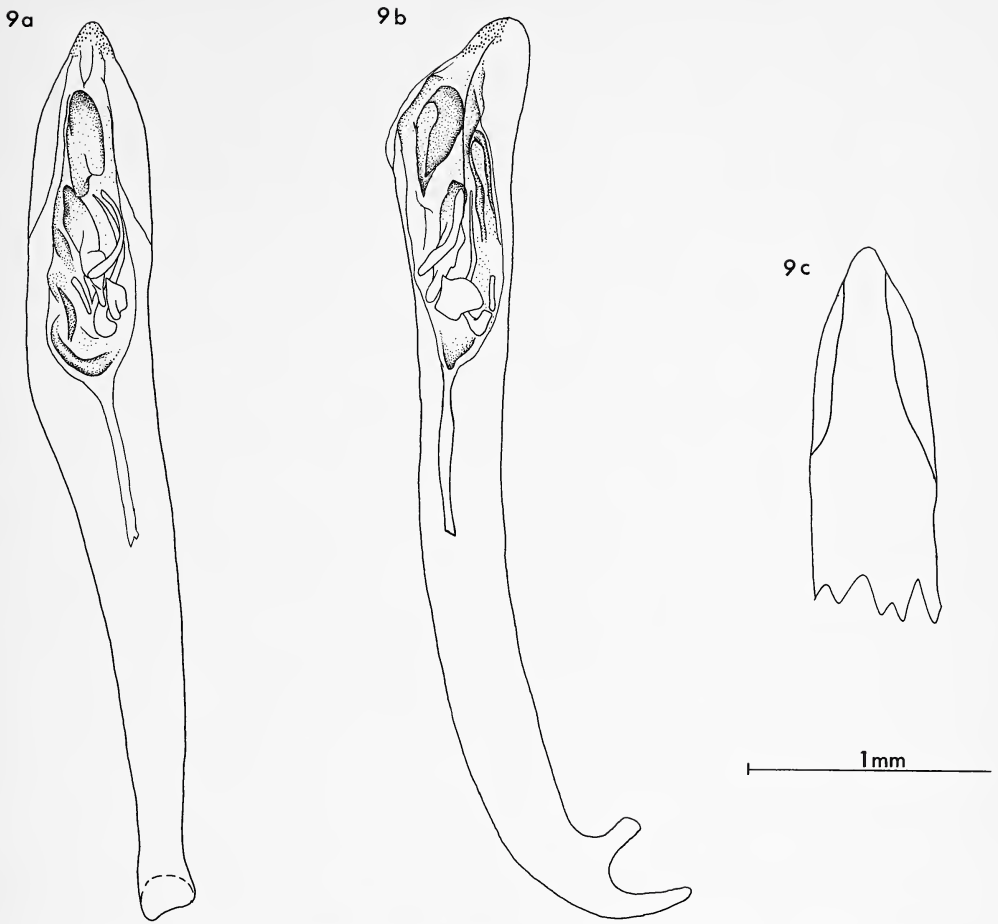
Fig. 9. Median lobe and inverted internal sac of *Cicindela theatina*. 9a, dorsal aspect; 9b, left lateral aspect; 9c, apical portion, dorsal aspect.

the male genitalia for grouping members of the *repanda* complex (*sensu* Leng) together with other species, which, I believe, should have been included in other species groups. The diagnosis of the North American species of the *maritima* group made by Rivalier (1954, Group IV) is followed here. Rivalier united members of the *repanda* group with *limbata* and *bellissima*, but *columbica* and *theatina* were not mentioned.

The following combination of characters of the internal sac of the male genitalia is regarded as being diagnostic, and separates the North American species of the *maritima* group from other species groups of *Cicindela* (see fig. 10): sclerite 1, a quadrate plate lying over the base of sclerite 4 (flagellum); sclerite 2, a flat, elongate, curved strip; sclerite 3, short, rectangular, and lying to the left of sclerite 4; sclerite 6, large, twisted basally, and lightly sclerotized apically; field *a* apical in the inverted position, roughened or densely aculeate; field *b*, a finger-like projection, roughened or densely aculeate; field *c*, semi-circular shape, terminal in the everted position; chitinous tooth (defined by Papp, 1952) absent.
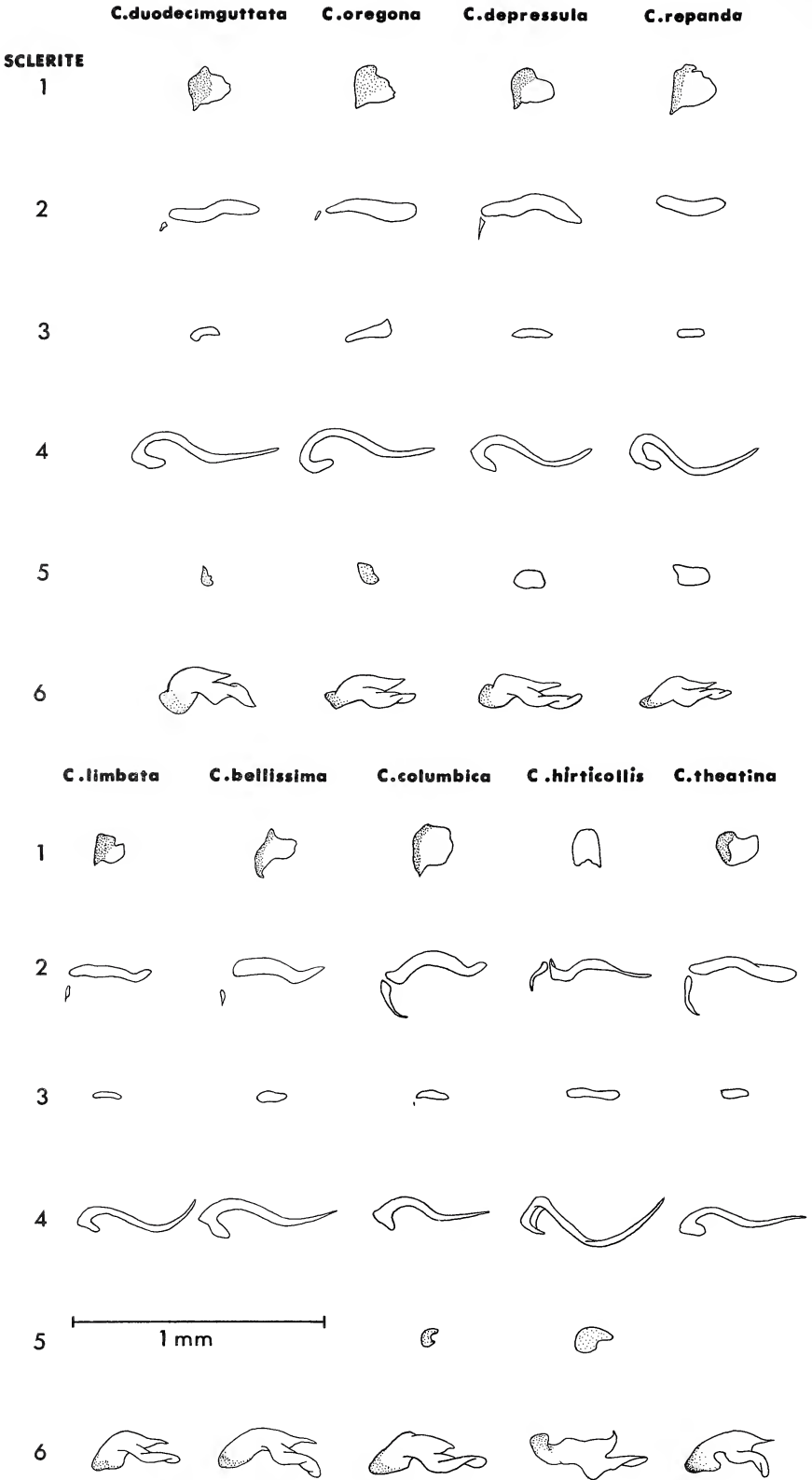
Fig. 10. Sclerites of the internal sac of the North American species of the *maritima* group, numbered as in fig. 1.

Keys to the North American Species of the *maritima* Group

Based on Non-genitalic Characters

For species marked *, reference to the median lobe of the male is advisable.

1   Genae glabrous....................................... 2
           Genae hairy (if hairs of the head or genae are abraded their
           former positions are indicated by tiny setigerous punctures .. 6
2 (1) Posterior tip of humeral lunule (when present) usually with
           a slight anteriorly-directed hook; head with frons covered
           with numerous hairs.........................*C. hirticollis* Say
           (not treated in detail).
           Posterior tip of humeral lunule not hooked; dorsum of head
           covered sparsely with hairs;OR glabrous;OR hairs clustered
           near the front inner edge of each eye...................... 3
3 (2) Marginal band of elytron absent.......................... 4
           Marginal band of elytron present......................... 5
4 (3) Cluster of one to four hairs near each eye; shoulder of
           middle band (usually) smoothly rounded;vertex often with
           several very small hairs........... * *C. depressula* Casey (p.   )
           Cluster of eight to 11 hairs near each eye; shoulder of
           middle band (usually) raised; vertex usually glabrous..........
           ....* *C. oregona* LeConte (p.   )
5 (3) Frons sparsely hairy; humeral lunule elongate and markedly
           oblique; posterior tip of humeral lunule nearly touching
           shoulder of middle band..................... *C. bellissima* Leng
           (not treated in detail).
           Frons glabrous; humeral lunule short and slightly oblique;
           posterior tip of humeral lunule widely separated from shoulder
           of middle band............................ *C. columbica* Hatch
           (not treated in detail).
6 (1) Humeral lunule c-shaped or in the form of two dots; elytral
           markings narrowly expanded or broken..................... 7
           Humeral lunule oblique; elytral markings very broad,
           widely connected, or brown pigment of elytra greatly
           reduced obliterating basic elytral markings................. 8
7 (6) Form broader than *repanda*; pronotum broad; marginal
           band absent or widely separated from humeral lunule..........
           .... * *C. duodecimguttata* Dejean (p.   )
           Pronotum narrow; marginal band connected to humeral
           lunule...................................* *C. repanda* Dejean
           (not treated in detail).
8 (6) Elytra predominantly pale, elytral pattern completely
           obliterated..................................... *C. limbata* Say
           (not treated in detail).
            Elytra predominantly dark............................... 9
9 (8) Marginal band broad and widely connected to other elytral

markings; posterior portion of humeral lunule short. . . . .
. . . . *C. theatina* Rotger
(not treated in detail).

Marginal band short connected only to middle band; posterior
portion of humeral lunule very long. . . . . *C. limbata* Say
(not treated in detail).

Based on the Male Genitalia

1   Median lobe with apical lateral flanges narrow. 2
         Apical lateral flanges of median lobe broad. 4
2   (1) Chamber of internal sac extended dorso-apically; sclerite
         between 2 and 6 large; sclerite 4 with a pronounced twist. . . . . .
         . . . . *C. hirticollis* Say (figs 8, 10)
         Chamber of internal sac not extended; sclerite between
         2 and 6 small or absent; twist in sclerite 4 normal. 3
3   (2) Sclerite 5 large; no sclerite between 2 and 6; part of
         median lobe containing internal sac short. . . . .
         . . . . *C. repanda* Dejean (figs 4, 10)
         Sclerite 5 normal size; sclerite between 2 and 6 small;
         part of median lobe containing internal sac elongate. . . . .
         . . . . *C. oregona* LeConte (figs 2, 10)
4   (1) Apical portion of median lobe with a distinct keel along
         median line. . . . . *C. theatina* Rotger (figs 9, 10)
         Keel absent. 5
5   (4) Apex of median lobe produced into a narrow tip. 6
         Apex of median lobe blunt, not produced. 7
6   (5) Sclerite 5 absent; fields *a* and *b* densely aculeate. . . . .
         . . . . *C. bellissima* Leng (figs 6, 10)
         Sclerite 5 normal size; fields *a* and *b* lightly aculeate. . . . .
         . . . . *C. duodecimguttata* Dejean (figs 1, 10)
7   (5) Lateral flanges of median lobe constricted basally; sclerite
         between 2 and 6 large. . . . . *C. columbica* Hatch (figs 7, 10)
         Lateral flanges of median lobe not constricted; sclerite between
         2 and 6 normal size. 8
8   (7) Sclerite 5 absent; fields *a* and *b* densely aculeate. . . . .
         . . . . *C. limbata* Say (figs 5, 10)
         Sclerite 5 present; fields *a* and *b* lightly aculeate. . . . .
         . . . . *C. depressula* Casey (figs 3, 10)

The Species *Cicindela duodecimguttata* **Dejean**

Cicindela duodecimguttata Dejean 1825:73. Type locality - Amerique
        septentrionale. Fall 1901:308. Leng 1902:148. Blatchley
        1910:34. Casey 1913:28. Horn 1915:374, and 1930:80.
        Stainer 1934:247. Papp 1952:515. Rivalier 1954:252.
        Lindroth 1955:16. Wallis 1961:20. Graves 1963:498.
        Cicindela bucolica Casey 1913:28. Type locality - Aweme,
        Manitoba. Wallis 1961:21.
        Cicindela hudsonica Casey 1916:29. Type locality - Hudson

Bay Territory. Wallis 1961:21.
Cicindela repanda edmontonensis Carr 1920:218. Type locality -
Edmonton, Alberta. NEW SYNONYMY
Cicindela repanda duodecimguttata, Horn 1930:81 (not Dejean);
Papp 1952:515.

This species is characterized by its dull brown dorsal surface,
and elytral maculations (see figs 11, 13, 15). Specimens of
duodecimguttata are usually distinguishable from specimens of the markedly
similar species *repanda*. In western areas, where these species are
sympatric, individuals of *duodecimguttata* have a broad prothorax, dark brown
elytra, and widely interrupted marginal bands, while specimens of *repanda*
have a narrower prothorax, lighter brown elytra, and the marginal bands
are narrowly, or not interrupted. In eastern Canada the elytral pattern
of *duodecimguttata* generally is broken but *repanda* retains full elytral macul-
ations excepting the subspecies *novascotiae* Vaurie that occurs on the
Canadian Atlantic coast (see Vaurie 1951). Differences in male genitalia
however, are clear and should be used for definitive identification (see
p. 91 & Lindroth 1955: 16-17).
       In western Canada, populations of *duodecimguttata* occur on the edges
of lakes, ponds, rivers, streams, and sloughs wherever the soil is dark
and wet and consists of mixtures of sand and clay, and clay or mud. This
type of habitat is preferred by *duodecimguttata* on the mainland in eastern
Canada (see Leng 1902, Blatchley 1910, and Graves 1963). Lindroth
(1955) in Newfoundland found *duodecimguttata* on sand and gravel, as well
as on clay or humus.

Notes on Synonymy

       The name *bucolica* Casey has been given to specimens of *duodecimguttata*
with full elytral markings. Such specimens are common in the western
prairies. Casey's *hudsonica*, the elytral pattern of which is very reduced,
is a variant of *duodecimguttata*. The name *edmontonensis* Carr was proposed for
a variant of *duodecimguttata* that has a narrow elytral pattern. Horn (1930)
treats *repanda* as a larger race of *duodecimguttata* and *bucolica* as a lesser form
of *repanda*. Evidence for this synonymy is presented in the following sec-
tion on geographic variation.

Geographic Variation

       This species inhabits a territory that extends from the eastern
front of the Rocky Mountains to the Atlantic seaboard, and from the North-
west Territories to Alabama (fig. 17). Throughout the range of
duodecimguttata, except for the zone of intergradation with *oregona*, two easily
observed characters vary geographically: color of the dorsal surface of
the body and elytral pattern. Variation in both features has been ex-
amined quantitatively. Variation in length of elytra has also been studied.
       Average lengths of elytra, from the tip of the scutellum to the end
of the elytral spine, were calculated for males and females from the 20
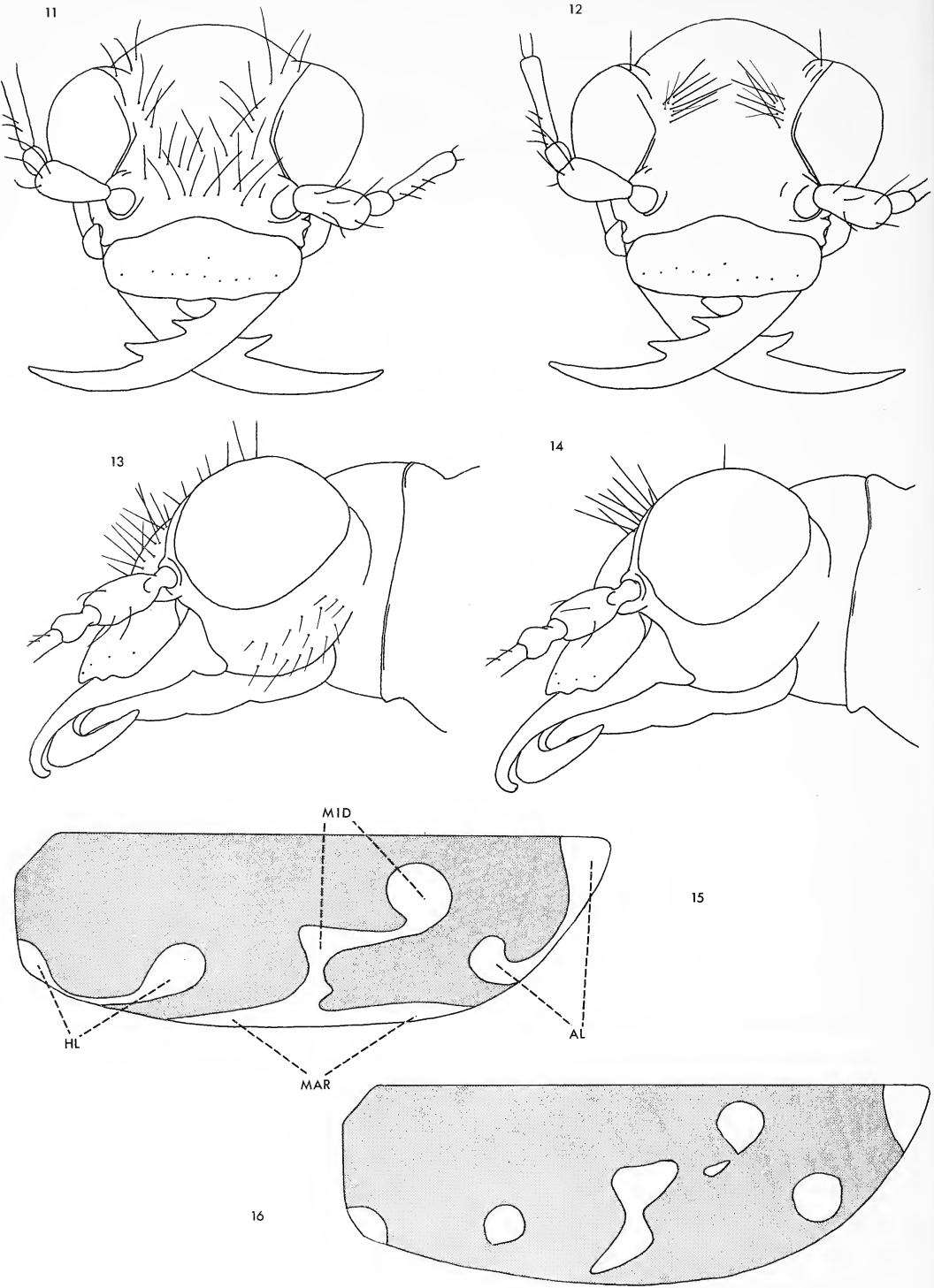
Figs 11 and 12. Anterior views of the heads of *C. duodecimguttata* and *C. oregona*. Figs 13 and 14. Lateral views of the heads of *C. duodecimguttata* and *C. oregona*. Figs 15 and 16. Left elytra of *C. duodecimguttata* and *C. oregona*: HL, humeral lunule; MAR, marginal band; MID, middle band; AL, apical lunule.

localities listed in table 3. Character gradients are irregular and do not conform with latitudinal, longitudinal, or altitudinal changes. The mean lengths of elytra range from 6.57 mm to 7.60 mm for males, and 7.11 mm to 8.22 mm for females. Average elytral lengths of males and females from the island of Newfoundland (Harmon Field) are not larger than those of corresponding sexes from coastal localities of the adjacent mainland such as Bathurst, New Brunswick.

There are no color differences between sexes of *duodecimguttata*. Males and females are usually dull brown dorsally, metallic blue-green ventrally, and the thoracic pleura are coppery. The dorsal surface is the only area that is subject to color variation. In most regions the dorsum is dull brown, but in eastern Canada and United States, color varies.

Specimens from seven localities on or near the Atlantic seaboard were examined for color of the dorsal surface; the results are listed in table 1. Brown specimens are most abundant in all of the population samples, followed in number by brown-green or green and finally blue individuals.

The most variable maritime population sample is one collected at Yarmouth, Nova Scotia. The entire color range is represented. Brown specimens account for 56 per cent of the sample. Green, brown-green, and blue individuals follow in number in that order. Blue specimens are absent from the Goose Bay, Labrador, and Harmon Field, Newfoundland samples both of which are composed mainly of brown members, followed by brown-green and green. Only brown individuals occur in the Bathurst, New Brunswick population sample. Except for one green specimen from Keene Valley, New York, the inland samples are made up entirely of brown specimens. Green specimens are not uncommon in coastal populations of species of the *maritima* group (see *o. oregona* and *o. novascotiae* Vaurie 1951).

The elytral pattern is fully developed in some *duodecimguttata* individuals and almost absent in others. The four main components of the elytral pattern vary independently. There were assigned numerical values to form a compound character index for analysing variation in elytral pattern. If all markings are complete a high score is assigned (maximum value 11), and if the markings are greatly reduced a low value is assigned (minimum value 0). Markings that range between complete and reduced are given intermediate values. The components of the elytral pattern are illustrated in fig. 15 and their assigned values are given in table 2. As many eastern *duodecimguttata* specimens have maculations typical of *oregona*, the compound character index used in this section was not employed in the hybridization section. A compound character index (hybrid index), based on the elytral pattern, was determined for each specimen of 20 population samples from different localities. Results are presented in table 3. The average index value for each sample is indicated in fig. 17. The samples are arrayed in five transects so that geographical variation in elytral maculations may be more clearly appreciated. Three transects - A-A', B-B', and C-C' - are run from west to east, while two - D-D', and E-E' - are oriented north to south.
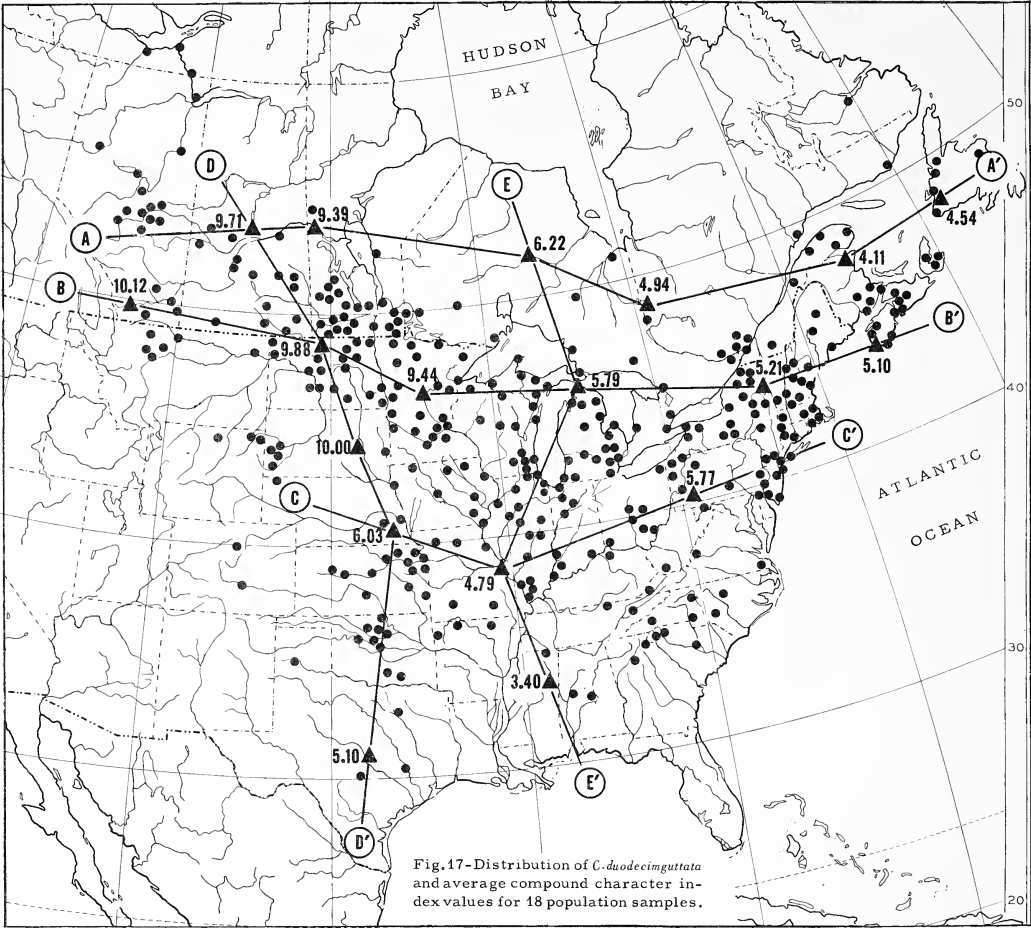
Fig.17-Distribution of *C.duodecimguttata* and average compound character index values for 18 population samples.

TABLE 1 - Variation in color of dorsal surface of *C. duodecimguttata* from seven eastern N. American localities.

Locality	No. Brown	No. Brown-green	No. Green	No. Blue	Total No.
Goose Bay, Labr.	16	12	1	0	29
Harmon Field, Nfld.	41	14	4	0	59
Bathurst, N. B.	19	0	0	0	19
Yarmouth, N. S.	29	8	11	4	52
Duparquet, Que.	17	0	0	0	17
Keene Valley, N. Y.	45	0	1	0	46
Jeannette, Pa.	31	0	0	0	31

TABLE 2 - Values assigned to elytral markings of *C. duodecimguttata* specimens for determination of compound character indices.

Elytral Markings	Values			
	0	1	2	3
Humeral lunule	1 dot	2 dots	broken	full
Middle band	1 dot	2 dots	broken	full
Apical lunule	1 dot	2 dots	broken	full
Marginal band	absent	trace	full	-

An average index reduction from west to east is seen in the A-A' transect. The mean values 9.73 and 9.39 of the samples that represent Christopher Lake, Saskatchewan and The Pas, Manitoba, respectively, indicate full elytral markings. The mean index change per mile between these localities is about 0.00160. The mean index for the population sample from Ogoki, Ontario is 6.22 which is a change of 0.00488 index units per mile from that of The Pas, Manitoba. The trend is less marked between Ogoki, Ontario and Duparquet, Quebec the rate of change being 0.00320 index units per mile. A change in average index value of 0.00133 occurs between Duparquet, Quebec (4.94) and Bathurst, New Brunswick (4.11), and a change of 0.00127 occurs between Bathurst, New Brunswick and Harmon Field, Newfoundland (4.54).

Average index values of the six population samples in transect B-B' complement the trend shown in A-A'. Elytral maculations are quite full in western localities as shown by average index values: Lethbridge, Alberta, 10.12; Bottineau County, North Dakota, 9.88; and Minnesota, 9.44. The rate of mean index change per mile between Lethbridge, Alberta and Bottineau County, North Dakota is only 0.00041 and increases slightly to 0.00130 between Bottineau County, North Dakota and Minnesota. However, the average index value for the Cheboygan, Michigan specimens is markedly less than that of the Minnesota sample, the rate of change being 0.00776 index units per mile. This is approximately six times the rate of change between Bottineau County, North Dakota and Minnesota. Mean index differences between eastern population samples are slight. Average values for areas follow: Cheboygan, 5.79; Keene Valley, New York, 5.21; and Yarmouth, Nova Scotia, 5.10, showing a reduction in the elytral pattern.

The southernmost west-east transect, C-C', comprises the following population samples and average index values: Bennett, Nebraska, 6.03; St. Louis, Missouri, 4.79; and Jeannette, Pennsylvania, 5.77. The mean index decreases 0.00355 units per mile between Bennett and St. Louis while between St. Louis and Jeannette there is a mean increase of 0.00158 units per mile.

Transect D-D' is oriented north to south near the western limits of the range of this species. The pattern of the elytra tends to increase

TABLE 3 - Frequency distribution of compound character index values of specimens of *C. duodecimguttata* from 20 localities.

Localities	Compound character index values											N
	1	2	3	4	5	6	7	8	9	10	11	
Fort Smith, N. W. T.			4	2	8	7	10	12	32	70	17	162
Lethbridge, Alta.				2	3	3	6		27	162	107	310
Christopher L., Sask.			1			1	2	2	6	33	12	57
The Pas, Manitoba					1	2	1		3	11	5	23
Bottineau County, N. D.					2		1	5	6	40	20	74
Minnesota					1	4	6	4	12	28	18	73
Wolsey, S. D.							1		1	11	44	17
Bennett, Nebraska			9	9	13	8	18	12	2	3		74
St. Louis, Mo.			3	8	4	1	2			1		19
Texas			3	6	5	2	1	2	1			20
Ogoki, Ontario		1	4	7	7	9	5	4	2	7		46
Cheboygan Co., Mich.			10	15	25	21	9	10	5		5	100
Oktibbeha Co., Miss.		1	61	12	2	4		1				81
Duparquet, Quebec	1		4	3		8		2				18
Keene Valley, N. Y.			8	12	9	5	3	1	3	1	1	43
Jeannette, Pa.			4	7	6	4	4	1	2	2	1	31
Goose Bay, Labr.		1		6	11	2	3	2	1	3		29
Bathurst, N. B.			8	6	3		1	1				19
Yarmouth, N. S.		1	4	15	15	10	3	2	1	1		52
Harmon Field, Nfld.		2	15	20	9	6	2	3	1		1	59

north to south in the first part of the transect as follows: Christopher Lake 9.71, Bottineau County 9.88, and Wolsey, South Dakota 10.00. The spatial mean change in index units between these population samples is negligible in contrast to that which occurs between Wolsey and Bennett (0.01498). The mean index value at Bennett is 6.03, and it is 5.10 for Texas. The three northern localities therefore have samples with full elytral markings, while specimens of the two more southerly localities have reduced maculations, a sharp change occurring between Wolsey and Bennett.

      A clinal north to south fragmentation in maculation of the elytra is evident in transect E-E'. Ogoki, Cheboygan, St. Louis, and Oktibbeha County, Mississippi have population samples with mean index values of 6.22, 5.79, 4.79, and 3.40 respectively. The rate of increase in mean index units per mile is 0.00101 between Ogoki and Cheboygan, 0.00170 between Cheboygan and St. Louis, and 0.00376 between St. Louis and Oktibbeha County, Mississippi.

      The population samples can be separated into two geographic groups by areas of marked rates of change in mean index values. The

greatest differences in average index values are between The Pas, Manitoba and Ogoki, Ontario; between Minnesota and Cheboygan County, Michigan;and between Wolsey, South Dakota and Bennett, Nebraska. The species therefore may be divided into northwestern populations that have complete elytral markings, and southern and eastern groups that exhibit a more or less interrupted elytral pattern. However, the two aggregates of populations are not subspecifically distinct. A separation on the basis of the 75 per cent rule cannot be made becaues of extensive overlap in range of variation between the two groups of populations.

Breakdown of the elytral pattern has probably occurred independently in *duodecimguttata*, *repanda, depressula*, and *oregona* and if so this is a good example of parallel evolution. Perhaps the breakdown of elytral pattern in *duodecimguttata* is the result of a mutation that has spread throughout most populations except for those in the west.

Canada. ALBERTA: Andrew, 3; Beaver Hill Lake, 1; Bilby, 14; Chin, 8; Cooking Lake, 7; Cypress Hills, 2; Doussal, 1; Drayton Valley, 4; Edmonton, 95; Falles, 1; Flatbush, 1; Fort Chipewyan, 1; Halfway House, 3; Jct. Rte. 39 and North Saskatchewan River, 11; Lake Cardinal, 2; Lesser Slave Lake (east end), 1; Lethbridge (St. Mary's River), 5; Lethbridge (Six-mile Coulee), 310; Louis Bull Reservation, 3; McMurray, 18; Medicine Hat, 5; mile 7 on Smith-Fitzgerald Road, 1; Redwater, 2; Stirling Lake, 1; Tilley, 1; Tofield, 19; Vilna, 1; Wabamun, 3. LABRADOR: Goose Bay, 29. MANITOBA: Aweme, 12; Baldur, 1; Beaver Lake, Riding Mountain, 2; Berens River, 9; Birtle, 1; Brandon (15 miles south), 2; Carberry (5 miles west), 1; Clear Lake, Riding Mountain, 2; Dauphin, 5; Delta, 3; Douglas, 1; Gladstone, 2; Glenboro, 1; Grunthal, 1; Hilton (6 miles south), 21; Holland, 2; Husavick, 3; Kelwood, 1; The Pas, 23; Magnuls, 1; Makinock, 5; Marchand, 1; Max Lake, Turtle Mountain, 10; Melita, 1; mile 360, Rte. 10, 1; Morris, 1; Ninette, 25; Norgate (5 miles west), 5; Oak Lake (4 miles west), 1; Red River, 1; Red Rock Lake, 1; Rennie (15 miles east), 1; Riding Mountain, 7; Rounthwaite, 1; Sandilands, 3; Shilo (5 miles south west), 4; Shoal Lake, 1; Silver Falls, 2; South Junction, 3; Stonewall, 1; Treesbank, Assiniboine River, 35; Vassar, 1; Victoria Beach, Lake Winnipeg, 8; Wanless, 1; Wasagaming, 1; Watson's Lake, 1; Waugh, 1; Westbourne, 5; Whitemouth, 1. NEW BRUNSWICK: Apohaqui, 1; Bathurst, 19; Chipman, 2; Penobsquis, 3; St. John, 1; Sackville,3; Shediac, 10. NEWFOUNDLAND: Bay of Islands, 4; Bay St. George, 13; Codnoy, 11; Deer Lake, Humber River, 2; Gander, 14; Harmon Field, 59. NORTHWEST TERRITORIES: Fort Smith, 163; Hay River (Great Slave Lake Shore), 1; Seven Mile Lake (26 miles west Fort Smith), 6; Resolution, 5. NOVA SCOTIA: Armdale, 3; Baddeck, 2; Barrington Passage, 1; Boisdale, 16; Cape Breton, 35; Cow Bay, 29; Bigby, 1; Great Village, 8; Halifax, 1; Ingramport, 5; Kedgemakooge Lake, 2; Kentville, 11; Lockeport, 2; North Sidney, 9; Port Maitland, 36; Queens, 2; Truro, 10; Weymouth, 1; Wilmot, 1; Yarmouth, 52. ONTARIO: Agawa Bay, Lake Superior, 1; Coniston, 3; DeCew Falls, 1; Goderich, 1; Gravenhurst, 1; Hamilton, 2; Hearst (65 miles west), 23; Hudson Bay, 1; Ingolf, 9; James Bay, 1; Kearney, 6; Kenora (14 miles east), 1; Lake of the Woods, Harris Hill, 2; Loleo, 1; Minnitaki, 1; Moose Factory, 6; Ogoki, 46; Ojibway, 1; One Sided Lake, 2; Ottawa, 2; Port Arthur, 3; Powasson, 2; Sibley Provincial Park, Middlebrun Bay, Lake Superior, 7; Sudbury, 1; Toronto, 5; Ventnor, 2; Woodtick Island, 1. QUEBEC: Baie Comeau, 1; Cap Rouge, 1; Cascapedia, 2; Charlevoix County, 4; Duchesnay, 6; Duparquet, 17; Gaspe, 5; Joliette, 2; Knowlton, 2; Lachute, 2; Ladysmith, 1; Lake Blanch, 12; Lac Opasatika, 1; Mont Joli, 2; Mont Lyall, 2; Montreal, 2; Natashquan, 1; Otter Lake, 12; St. Alexandre, 1; Ste. Anne de Monts, 1; Val Morin, 1. SASKATCHEWAN: Big River, 16; Broadview, 4; Candle Lake, 3; Ceylon, 1; Christopher Lake, 24; Cut Knife, 4; Estevan, 2; Fish Lake, 1; Glaslyn, 3; Good Spirit, 10; Holbein, 1; Kenosee, 11; Lake Manitou, 1; Neat Frys, 9; Pike Lake, 1; Pike Lake Park, 41; Regina, 1; Saskatoon, 22; Val Marie, 2; Waskisiu Lake, 10; White Fox, 12; Yorkton, 2.

United States. ALABAMA: Chilton County: Coosa River, 1. Tuscaloosa County, 1. ARKANSAS: Boone County: Harrison, 2. Lawrence County, 1. Washington County: Fagett, 1. Localities of unknown counties: Ozark Mountains, 11. COLORADO: Fremont County: Coal Creek, 1. CONNECTICUT: Litchfield County: Cornwall, 3; Litchfield, 2; Torrington, 3; Twin Lakes, 4; Washington, 2. New Haven County: Meriden, 7. DELAWARE: New Castle County: Newark, 6. GEORGIA: Fulton County: Atlanta, 8; East Point, 1. Habersham County: Cornelia, 1. Localities of unknown counties: Georgia, 1. ILLINOIS: Champaign County: Champaign, 1; Urbana, 4. Cook County: Chicago, 17; Cook County, 2; Flossmoor, 3; Palos Park, 2; Summit, 10. Fayette County: Ramsey, 2. Lake County: Antioch, 1; Cedar Lake, 3; Lake County, 1; Waukegan, 3. McHenry County: Algonquin, 3; McHenry, 1; Richmond, 3. McLean County: Bloomington, 1; Normal, 2. Macon County: 1. Marshall County: Taluca, 1. Ogle County: White Pines Forest, 6. Peoria County: Peoria, 4. Perry County: Pyem, 1. Piatt County, Atwood, 1. Putnam County: 2. Randolph County: Chester, 3. Rock Island County: Moline, 1. Union County: Ware, 3. Will County: New Lenox, 1. Williamson County: Crab Orchard Lake, 1. Localities of unknown counties: Dune Park, 1. Edgebrook, 16. Funks Grove, 4. Illinois, 1. Rock, 1. INDIANA: Cass County: 1. Gibson County: 2. Jefferson County: Hanover, 1. Knox County: Vincennes, 1. Porter County: Beverley Shores, 1. Posey County: 3. Starke County: North Judson, 12. Tippecanoe County: Lafayette, 3. Localities of unknown counties: Lake Station, 1. Mineral Springs, 3. Pine, 1. T.R.P. Indiana, 1. IOWA: Boone County: Boone, 27. Cerro Gordo County: Clear Lake, 1. Clayton County: Guttenberg, 1. Dickinson County: Lake Okoboji, 3. Henry County: Mount Pleasant, 1. Howard County: Elma, 1. Johnson County: Iowa City, 4. Lee County: Fort Madison,1. Story County: Ames, 2. Woodbury County: Sioux City, 1. Localities of unknown counties: Iowa, 1. Silver Lake, 1. KANSAS: Atchison County: Atchison, 3. Bourbon County: Fort Scott, 3. Coffey County: Burlington, 1. Douglas County: 3. Ellis County: 1; Hays, 1. Johnson County: Argentine, 14. Leavenworth County: Leavenworth, 7. Pottawatomie County: Onaga, 8. Riley County: 1. Saline County: Salina, 1. Shawnee County: Topeka, 11. Trego County: Wakeenay, 1. Localities of unknown counties: Central Kansas, 1. KENTUCKY: Localities of unknown counties: Kentucky, 3; Kentucky near Cincinnati, Ohio, 1. Maine: Hancock County: Bar Harbor, 1; Mount Desert, 6; Seal Harbor, 7. Kennebec County: Monmouth, 12. Piscataguis County: Greenville, 1; Mount Katahdin, 1. York County: Agamenticus, 2. Localities of unknown

counties: Bass Harbor, 2; Maine, 1; Pleasant Ridge, 5; Wales, 2. MARYLAND: Allegheny County: Mount Savage, 4. MASSACHUSETTS: Berkshire County: Benedict Pond, 2; Lenox, 4. Bristol County: Rehoboth, 2. Middlesex County: Framingham, 19; Sherborn, 9. Norfolk County: Sharon, 1. Plymouth County: 3. Suffolk County: Cambridge, 1; Medford, 1; Stoneham, 1. Worcester County: Southboro, 2. MICHIGAN: Alger County: 2; Onota Twp., 10. Allegan County: Allegan State Forest, 1; Rabbit River, 1. Alpena County: Alpena, 1; Squaw Bay, 2. Arenac County: White Stone Point, 1. Berrien County: Sawyer Dunes, 1. Cass County: 1. Charlevoix County: Beaver Island, 1; Thumb Lake, 1. Cheboygan County: 16; Douglas Lake, 84. Chippewa County: Marquette N.F., 1; Neebish Island, 1. Clare County: 8-Point Lake, 1. Delta County: Garden, 1. Genesee County: Davison T.W.P., 1; Flint, 1. Gogebic County: 12; Black River Park, 4. Huron County: Pte. Aux Barques, 2; Port Austin, 1; Sand Point, 1. Ingham County: 1. Ionia County, 1. Iosco County: 2; State Game Refuge, 1. Keweenaw County: Copper Harbor, 1; Eagle Harbor, Lake Superior, 10; Manganese Lake, 11. Lapeer County: Hadley T.W.P., 1. Mackinac County: 1; St. Ignace, 3. Marquette County: Huron Mountains, 12; Marquette, 1. Menominee County: Daggett, 1. Monroe County: Erie, 1. Montmorency County: 3. Ontonagan County: Gogebic Lake, 15. Otsego County: 2; Vanderbilt, 1. Schoolcraft County: Germfask, 1. Tuscola County: Bay Park, 2. Wayne County: Detroit, 6. Washtenaw County: Ann Arbor, 12. Localities of unknown counties: Pcn. Ind., 1; Michigan, 2. MINNESOTA: Aitkin County: Aitkin, 1. Anoka County: 2. Becker County: 5. Beltrami County: Waskish, 3. Carlton County: Moose Lake, 3. Carver County: Lake Waconia, 2. Clearwater County: 14; Gonvick, 1. Hennepin County: 1; Minneapolis, 2. Koochiching County: Rainy Lake, 3. Lac Qui Parle County: 3. Lake County: South Kawishiwi, 1. Lake of the Woods County: Williams, 1. Le Sueur County: 1. Nicollet County: St. Peter, 5. Renville County: Bird Island, 1. Roseau County: Roseau, 2. St. Louis County: Duluth, 2. Scott County: Jordan, 1. Stearns County: Koronis Lake, 6; Rice Lake, 10. Traverse County: 3. Wilkin County: Rothsay, 1. MISSISSIPPI: Oktibbeha County: 12; A & M College, 72. Tippah County: Tiplersville, 2. MISSOURI: Caldwell County: Hamilton, 13. Carter County: Van Buren, Ozarks Mountains, 2. Greene County: Springfield, 2; Willard, 8. Linn County: 1. Pike County: Louisiana, 7. St. Louis County: St. Louis, 19; Valley Park, 2. MONTANA: Cascade County: Ulm, 1. Chouteau County: Fort Benton, 1. Hill County: Fresno, 2. Roosevelt County: Brocton, 1. Teton County: Chouteau, 3. Toole County: Dunkirk (8 miles south), 5. NEBRASKA: Dawes County: Wayside, 5. Lancaster County: Bennet, 73; Lincoln, 25; Malcolm, 31. Sarpy County: Bellevue, 1. NEW HAMPSHIRE: Caroll County: Ellis River Road, Jackson, 1; Wildcat Bank, Jackson, 1. Cheshire County: Jaffrey, 2. Coos County: Gorham, Peabody River, 4; Jefferson, 25; Israel River, Jefferson, 10. Grafton County: Twin Mountain, 21. Hillsboro County: Antrim, 1. Sullivan County: Meriden, 12. Localities of unknown counties: Glen House, White Mountains, 2; Martin Loe'n, White Mountains, 3; New Hampshire, 4; White Mountains, 1. NEW JERSEY: Bergen County: Ramsey, 1. Cape May County: Ocean City, 1. Essex County: South Orange, 1. Hudson County: Arlington, 6; Snake Hill, 5. Middlesex County: Jamesburg, 10; Milltown, 1. Passaic County: Paterson, 1. Salem County: Canton, 2. Sussex County: Lake Hopatcong, 1. Localities of unknown counties: Frieses Mill, 1; Manchester, 1; New Jersey, 2. NEW YORK: Cortland County: McLean Bogs, 1. Delaware County: Stamford, 2. Erie County: Buffalo, 1. Ebenezer, 1. Essex County: Ausable Lakes, 1; Elizabeth Town, 1; Heart Lake, 5; Jay Mountains, 1; Keene Valley, 46; Lake Golden, 1; Mount Whiteface, 3; Wilmington, 4. Franklyn County: Duane, 1. Fulton County: 1. Genesee County: Bergen, 6. Hamilton County: Lake Pleasant, 4; Racquet Lake, 3. Nassau County: Freeport, 1. New York County: New York City, 5. Niagra County: Lockport, 1. Onondaga County: White Lake, 5. Orangelo County: Pine Island, 3. Oswego County: Minetto, 2. Queen's County: Far Rockaway, 1. Richmond County: Clover Valley, Staten Island, 1; Hugenot, Staten Island, 1. St. Lawrence County: Cranberry Lake, 2. Tompkins County: Ithaca, 16. Warren County: Lake George, 1; Stamford, 3. Localities of unknown counties: Big Island, 3; Clearwater, 1; Luzerne, 1; Quaker Hill, 2. NORTH CAROLINA: Buncombe County: Black Mountains, 1. Guilford County: Jamestown, 1. Mecklenburg County: Charlotte, 2. Moore County: Manly, 2; Southern Pines, 2. Orange County: Chapel Hill, 2. Localities of unknown counties: Morrison, 6. NORTH DAKOTA: Benson County: 2. Bottineau County: 73; Lake Metigoshe, Turtle Mountain, 8; Omemee, 3. Burleigh County: 3; Bismark, 1. Burke County: 4. Cass County: Fargo, 1. Cavalier County: 4. Divide County: 4. Eddy County: New Rockford, 14; Sheyenne River, 3. Kidder County: Tappen, 1. Logan County: 4. McHenry County: 14. McLean County: 1. Morton County: 3. Nelson County: 4; Stump Lake, 3. Pembina County: 1. Ransom County: 3. Rawsey County: 1. Renville County: 8. Richland County: 2. Rolette County: 10; Golden Lake, Turtle Mountain, 2. Sheridan County: 2. Ward County: 4. Williams County: 3. Localities of unknown counties: Jarves Lake, 1; Mooreton, 3. OHIO: Ashtabula County: Ashtabula, 2; Jefferson, 17. Delaware County: 1. Franklin County: Columbus, 10. Gallia County: Vinton, 3. Hamilton County: Cincinnati, 4. Hocking County: 1. Licking County: Bowling Green Trail, 4; Newark, 1. Localities of unknown counties: Crane Hollow, 1. OKLAHOMA: Blaine County: Roman Nose State Park, 1. Cleveland County: Norman, 2. Garfield County: Enid, 1. Johnston County: 1. Kay County: Newkirk, 2. Kingfisher County: Kingfisher, 1. Murray County: Sulphur, 5. Noble County: Perry, 7. Payne County: Lake Carl Blackwell, 1; Stillwater, 1. Localities of unknown counties: Blue Jacket, 3; Centralia, 1; Wyandotte, 1. PENNSYLVANIA: Allegheny County: 5; Fair Oaks, 2; Westview, 1. Crawford County: Meadville, 1. Cumberland County: Mount Holly, 1. Delaware County: 4; Lansdowne, 4. Forest County: Endeavor, 1. Mercer County: Sharpsville, 1. Montgomery County: 1. Philadelphia County: Lawndale, 1; Philadelphia, 3. Warren County: Bear Lake, 1. Westmoreland County: Jeannette, 31. Localities of unknown counties: Castle Rock, 7; Pennsylvania, 1; Springfield, 2; Wali, 1. RHODE ISLAND: Providence County: Elmwood, 5. Washington County: Misquamicut, 2. SOUTH CAROLINA: Greenville County: Greenville, 1. Pickens County: Clemson College, 5. Richland County: Columbia, 11. SOUTH DAKOTA: Beadle County: Wolsey, 17. Fall River County: Hot Springs (5 miles south), 1. Lawrence County: Deadwood, 1; Savoy, 2. Meade County: Sturgis, 3. Moody County: Colman, 2. Pennington County: Rapid City, 1. Localities of unknown counties: South Dakota, 1. TENNESSEE: Knox County: Knoxville, 2. Pickett County: 1. TEXAS: Blanco County: 1. Dallas County: 1; Dallas, 10. Randall County: Canyon, 1. Washington County: Burton, 2. Localities of unknown counties: Cyp. Mills, 1; Texas, 4. VERMONT: Bennington County: Mount Equinox, 2. Localities of unknown counties: Vermont, 1. VIRGINIA: Bath County: Warm Springs, 4. Lee County: Pennington Gap, 1. Nansemond County: Suffolk, 2. Localities of unknown counties: Black Pond, 1; Virginia, 1. WEST VIRGINIA: Wyoming County: Pineville, 3. WISCONSIN: Bayfield County: Lake Namekagon, 1. Clark County: Wordon Township, 1. Dane County: 2. Dodge County: Beaver Dam, 10. Douglas County: Superior, 1. Kewaunee County: Kewaunee, 1. Milwaukee County: Milwaukee, 1. Vilas County: Tenderfoot Lake, 1. Walworth County: Allens Grove, 2. Waukesha County: Oconomowoc, 1. Waupaca County: Waupaca, 1. Wood County: Cranmoor, 4. Localities of unknown counties: Walker, 1; Wisconsin, 1. WYOMING: Crook County: Alva (6 miles east), 3; Devil's Tower, 6. Sheridan County: Sheridan (8 miles north west), 4.

The Species *Cicindela oregona* **LeConte**

Cicindela oregona oregona LeConte 1857:41. Type locality - Oregon Territory
and northern California as far as San Francisco. Fall 1901:308.
Leng 1902:149. Casey 1913:29. Horn 1915:377, and 1930:82.
Varas Arangua 1928:247. Tanner 1929:83. Papp 1952:514. Hatch
1953:41 (see Hatch 1953 for more references to *o. oregona*).
Cazier 1954:242. Rivalier 1954:252. Wallis 1961:22.
  Cicindela guttifera , Fall 1901:308.
  Cicindela guttifera , Leng 1902:150.
  Cicindela depressula scapularis Casey 1909:272. Type locality-California.
  Cicindela guttifera sonoma Casey 1913:29. Type locality - California
  (maritime regions north of San Francisco). Horn 1915:378.
  Cicindela quadripennis Casey 1913:30. Type locality - Hawthorne,
  Nevada. Horn 1915:378.
  Cicindela ovalipennis Casey 1913:30. Type locality - Hawthorne,
  Nevada. Horn 1915:378.
  Cicindela oregona scapularis, Horn 1915:378.
Cicindela oregona guttifera LeConte 1857:42. Type locality - New Mexico.
Leng 1902:41. Horn 1915:378, and 1930:82. Varas Arangua 1928:
250. Tanner 1929:83. Cazier 1954:242. Wallis 1961:22.
  Cicindela sterope Casey 1913:28. Type locality - Kansas. Horn 1915:
  378.
  Cicindela audax Casey 1913:29. Type locality - Colorado. Horn 1915:
  378.
  Cicindela guttifera , Casey 1913:29.
  Cicindela oregona oregonella Casey 1924:16. Type locality - Parowan,
  Utah.
  Cicindela duodecimguttata, Hatch 1953:38 (not Dejean).
Cicindela oregona guttifera x *Cicindela oregona maricopa*
  Cicindela provensis Casey 1924:15. Type locality - Parowan and Provo
  Canyon, Utah.
  Cicindela provensis mormonella Casey 1924:15. Type locality - Eureka,
  Provo Canyon, Parowan and Vineyard, Utah.
  Cicindela provensis nephiana Casey 1924:16. Type locality - Parowan,
  Utah.
  Cicindela oregona maricopa, Tanner 1929:83 (not Leng).
Cicindela oregona maricopa Leng 1902:150. Type locality - Phoenix, Arizona.
Horn 1915:378. Horn 1930:82.
  Cicindela guttifera maricopa , Casey 1913:27. Varas Arangua 1928:250.
Cicindela oregona navajoensis Van Dyke 1947:155. Type locality - Kayenta,
Arizona.

On the basis of a patch of hairs confined to the front inner edge of
each eye this species may be distinguished from all other tiger beetles of
the *maritima* group, except *depressula* and female *scutellaris* Say (see figs 12,
14, 16). Female *scutellaris* and *oregona* specimens usually can be distinguish-

ed from each other simply by noting the geographical location from which the specimens were taken. The range of *scutellaris* is east of the Rocky Mountains while *oregona* occurs in the west limited by the eastern foothills of the Rockies.   Specimens of the subspecies *scutellaris scutellaris* are present in Colorado and New Mexico (Shelford 1917) but these forms are quite different from *oregona* in that they have bright cupreous to red elytra. Another subspecies of *scutellaris*, related to the subspecies *criddlei* , also occurs in Colorado (Rumpp 1961) and it is characterized by broad white margins of the elytra.   The species *depressula* and *oregona*, on the other hand are sympatric.   Individuals of these two species can be told apart by the numbers of hairs forming the clusters near each eye. The species *oregona* normally has eight to eleven hairs in this area while *depressula* usually has one to three and rarely four. A more reliable character for distinguishing between *oregona* and *depressula* is the shape of the median lobe of the male.

Like most other species of the *maritima* group *oregona* lives along the edges of rivers, lakes, and sloughs and is found on a variety of substrates. I have taken *oregona* on sandy beaches, gravelly banks, and indeed on rock. This species is more common where there are open patches of beach.

Notes on Synonymy

Casey proposed the names *C. quadripennis* and *C. ovalipennis* for male and female *C. o. oregona* respectively, that occur in Hawthorne, Nevada. Similarly *C. oregona* specimens collected north of San Francisco, were regarded by Casey as a subspecies of *guttifera* and he applied the name *sonoma* to them.   Casey also considered a coastal blue form of *o. oregona* to be a subspecies of *depressula* and named it *scapularis* . However *scapularis* does not itself occur in uniform geographic populations and consequently I have not given it taxonomic status (see Wallis 1961).

Casey's *audax* and *sterope* are both forms of *o. guttifera* . Their original descriptions indicate these names refer to typical *guttifera* in Colorado and New Mexico. The name *o. oregonella* Casey has been given to specimens from highly variable populations of *o. guttifera* which occur in north central Utah. *C. provensis* Casey refers to blue specimens that were taken in Parowan and Provo Canyon, Utah. Parowan is located in southwestern Utah, a hybrid area of *o. guttifera* and *o. maricopa* , and Provo Canyon is situated in north central Utah where *o. oregona* and *o. guttifera* intergrade. The name *provensis* represents hybrid individuals of these regions. Tanner regarded *guttifera* x *maricopa* and *guttifera* x *oregona* hybrid specimens in Utah as being variants of *o. maricopa*.

Geographic Variation and Subspecies

The species *Cicindela oregona* ranges widely in the west, from Alaska to southwestern California, Arizona, and New Mexico and eastward to the Rocky Mountains (fig. 18). Five easily observed characters vary geographically: body size, color of thoracic pleura, color of elytra, color of pronotum, and expanse of elytral pattern. Length of elytra is expressed in millimeters from the tip of the scutellum posteriorly to the

tip of the elytral spine along the suture. Width of elytron is similarly expressed in millimeters from the median line of the elytra through the transverse portion of the middle band to the elytral margin. These data are listed in tables 4 and 5 for males and females respectively. The tables also summarize data on variation in diameters of apical dots. The measurements illustrate variation in expanse of the elytral pattern. The apical dot was measured transversely across the widest portion.

Size - Before discussing the geographical aspects of size variation, I would note that females on the average are larger than males of the same population in every locality listed in tables 4, and 5. This is true for the sexes in the same locality, but is not necessarily true if opposite sexes of different regions are compared. For example, females from Trinidad, Colorado have a mean elytral length of 7.23 mm while the average elytral length of males from Tanana River, Alaska is 7.26 mm. Data on variation of elytral length in males and females are given in tables 4 and 5 respectively.

Three geographical routes (A, B, and C in column 1 of tables 4 and 5) have been selected to facilitate description of geographical variation in the length and width of elytra and expanse of color pattern. Tanana River, Alaska and Terrace and Oliver, British Columbia serve as the northern portion for all three routes. The first transect of population samples (A) extends from Alaska, south to New Mexico through British Columbia, Montana, Wyoming and Colorado. A second line of samples (B) is from Alaska to Arizona by way of British Columbia, Idaho and Utah. A third course (C) is from Tanana River, Alaska to southern Nevada, through British Columbia and Idaho. The data in tables 4 and 5 are arranged to correspond to these routes.

Because the corresponding character gradients of males and females are generally parallel, only the male samples are discussed in detail, with occasional reference to female samples. Table 4 indicates a decrease in the mean length of elytra of males, from north to south for all three courses. Each cline is quite irregular and there are sharp decreases and increases throughout. These abrupt changes in the character gradients appear to be correlated, at least in part, with changes in altitude or with geographic barriers. However, I have noted discrepancies in the clines that cannot be so related.

Through the northern section of the first route there is a southward decrease in average length of elytra of 0.007 mm per degree of latitude for males and 0.018 mm per degree of latitude for females. From Alaska to Lower Medicine Lake, Montana a distance of 14°30' of latitude, no marked deviations occur in the trend. Between Lower Medicine Lake and Hardy, Montana however, a distance of only 1°10' latitude, mean length decreases by 0.20 mm. There is a drop of 1,500 feet in altitude between these two localities. Another irregularity in the above character gradient occurs between Helena and Gardiner, Montana - an increase in mean length of elytra of 0.18 mm with 1°30' of latitude. Gardiner is 1,640 feet higher than Helena and contrast in elevation again seems to be related to the clinal difference. Population samples from Gardiner, Montana, Yellowstone National Park, Jackson Hole National Monument, and Moran, Wyoming have elytra of approx-
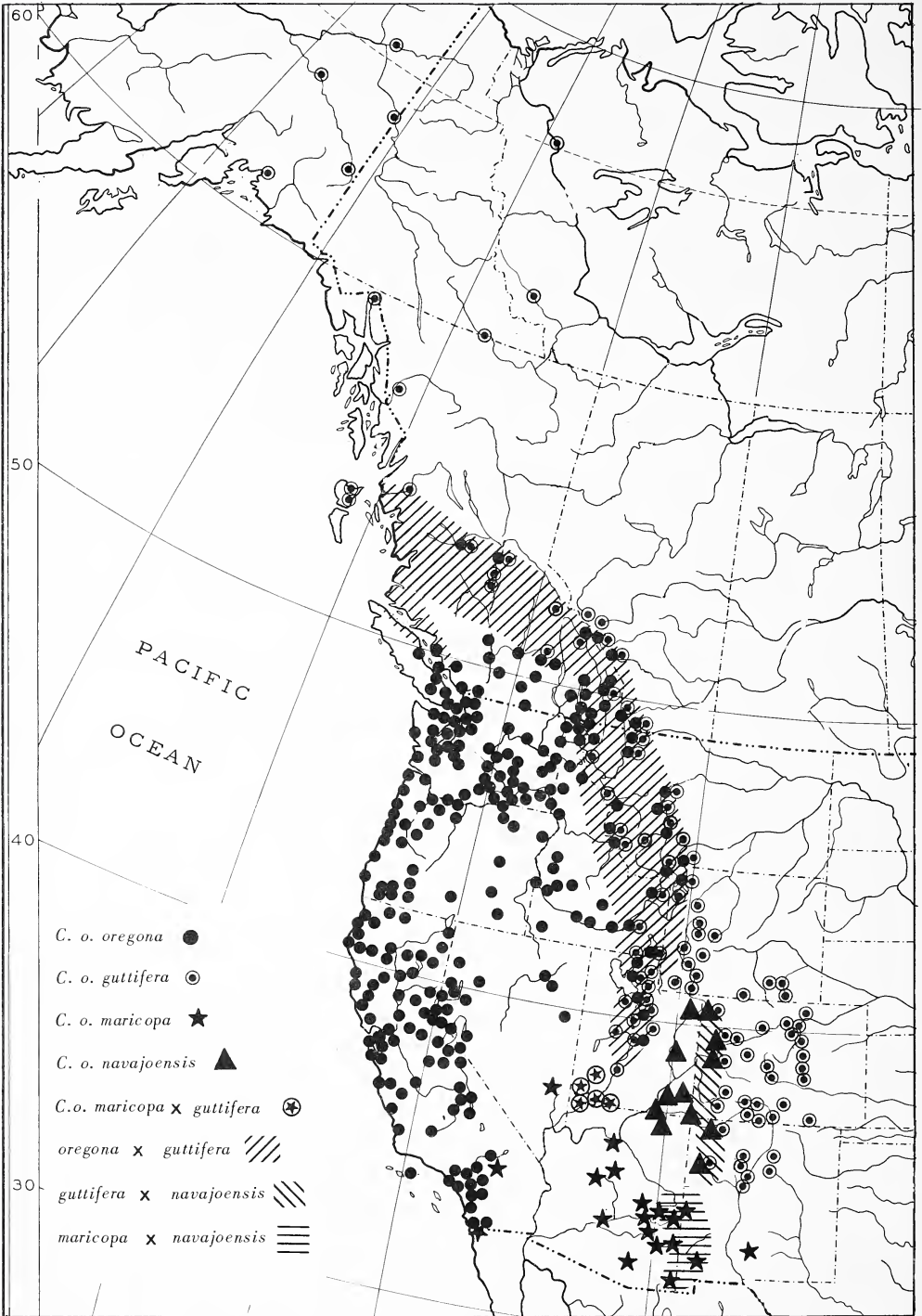
Fig. 18 - Geographical distribution of the subspecies of *C. oregona*.

imately equal length. Immediately southward the slope of the character gradient decreases markedly between Moran and Labarge, Wyoming. Both sites are at approximately the same altitude and there are no obvious geographic barriers between the two localities. In the Labarge and Green River regions the reduction in body size may be due to local factors such as disease, lack of food or marginal habitats (Mayr 1963). Jelm and Saratoga in western Wyoming are rather isolated from Fort Bridger and Green River, in eastern Wyoming by the Great Divide Basin and the Continental Divide which are situated in south central Wyoming. East to west gene flow between populations of *Cicindela oregona* is most likely impeded in southern Wyoming by these geographical features which may account for the shorter elytra in eastern Wyoming. The difference between the average elytral lengths of males from Jelm and Fort Bridger is statistically significant but that between females is not.

The second arbitrary line of population samples (B) is from Alaska to Arizona by way of British Columbia, Idaho, and Utah. A clinal decrease in length of elytra is evident throughout this route as well. In Utah, the Alta, Mount Timpanogos, and Provo Canyon populations have relatively long elytra. The elevation of Alta is 8,585 feet, Mount Timpanogos is 11,750 feet, and Provo Canyon is located in Provo Park which rises at a height of 11,068 feet. Samples collected at lower elevations in areas adjacent to the above mentioned, have a shorter mean elytral length and populations from Stockton and Provo are examples of these. Population samples taken in Salt Lake City may have been collected in any of the creeks entering the city from the Wasatch range which serves as the eastern geographic limits of the metropolis. Although the insects were labelled as being collected in Salt Lake City, they could conceivably have been taken at a much higher altitude nearby. Floy, Utah, and Kayenta, Arizona have populations with the shortest elytra in the entire span of this gradient. South of Kayenta the samples taken in Prescott, Phoenix, and Globe, Arizona are larger and compare in size with those from Idaho. These large forms in central and southern Arizona are fairly isolated and common only in these areas (see *oregona maricopa* p. 127).

The third line of population samples (C) extends from Alaska, south through British Columbia and Idaho to Nevada. Even though elytra are generally shorter in more southerly latitudes, the Walker Lake, Nevada population sample has the value for mean length of elytra equal to that of Tanana River, Alaska. I cannot account for this discrepancy.

Data on the variation in width of elytra are presented for males in table 4 and for females in table 5. There is a slight decrease in width of the elytra from Alaska, southward along all three routes. Irregularities in the clines of elytral widths correspond with changes in the character gradients of the lengths of elytra. At higher elevations of Montana, Wyoming and Utah mean values for elytral width are generally slightly greater than those of Alaska and Terrace, British Columbia. Tiger beetles of this species living in these lower latitudes at high altitudes are normally shorter but wider than their counterparts in boreal areas. This is especially marked in females. For example compare population samples of Tanana River, Alaska, Terrace and Queen Charlotte Islands, British Columbia with Gardiner, Montana, Alta and Provo Canyon, Utah,

TABLE 4 - Variation in male *Cicindela oregona.*

Length of Elytra

Route	Locality	North lat.	Elev. ft.	N	Range mm	$\overline{X} \pm$ SE	SD	CV
A, B, C	Alaska							
	Tanana R.	63.00	1500	16	6.65-7.73	7.26±0.09	0.35	4.75
A, B, C	British Columbia							
	Terrace	54.31	223	19	6.70-7.60	7.24 0.06	0.25	3.49
	Q. Ch. Islands	53.00	0-4100	7	6.81-7.70	7.34 -	-	-
	Oliver	49.10	2143	36	6.35-7.92	7.13 0.06	0.36	5.02
A	Montana							
	Low. Medicine							
	Lake	48.30	5000	44	6.91-7.62	7.16 0.03	0.18	2.49
	Hardy	47.11	3500	17	6.28-7.38	6.96 0.08	0.33	4.80
	Helena	46.35	4160	28	6.61-7.50	7.06 0.05	0.26	3.71
	Gardiner	45.03	5800	40	6.27-7.85	7.24 0.05	0.32	4.36
A	Wyoming							
	Yellowstone							
	Nat. Park	44.30	7000	77	6.13-7.98	7.19 0.04	0.38	5.33
	Jackson Hole							
	Nat. Mon.	43.50	6800	19	6.21-7.79	7.23 0.08	0.34	4.67
	Moran	43.52	6800	27	6.52-8.13	7.22 0.05	0.26	3.57
	11 miles S.							
	Labarge	42.15	6600	16	6.06-7.63	7.01 0.11	0.42	6.05
	27 miles S.							
	Labarge	42.14	6600	14	6.59-7.65	7.03 0.08	0.29	4.18
	Green River	41.33	6100	30	6.13-7.44	6.91 0.07	0.37	5.31
	Fort Bridger	41.19	6657	38	6.46-7.77	7.21 0.05	0.31	4.30
	Saratoga	41.28	6790	15	6.75-7.54	7.09 0.06	0.23	3.23
	Jelm	41.03	7500	46	6.21-7.66	7.01 0.04	0.28	3.95
A	Colorado							
	Estes Park	40.24	7547	12	6.32.7.32	7.02 0.08	0.27	3.85
	Trinidad	37.11	5999	16	6.23-7.03	6.69 0.06	0.25	3.75
A	New Mexico							
	Jemez Springs	35.45	6195	78	6.10-7.45	6.86 0.03	0.29	4.23
	Pecos &							
	Beulah	35.34	7000	10	6.40-7.18	6.73 0.09	0.27	4.01
	Fort Wingate	35.30	6997	13	6.25-7.10	6.66 0.08	0.27	4.05
B, C	Idaho							
	Valley County	45.00	-	7	6.28-7.56	7.01 -	-	-
	Owyhee County	42.30	-	22	6.15-7.61	7.03 0.09	0.40	5.66
	Bear Lake	42.05	5924	61	6.23-7.92	7.02 0.05	0.38	5.34
B	Utah							
	Ogden	41.14	4296	10	6.40-7.75	7.04 0.14	0.43	6.15
	Salt Lake City	40.45	4354	20	6.50-7.73	7.26 0.08	0.35	4.82
	Alta	40.36	8585	2	7.34-7.55	7.45 -	-	-
	Stockton	40.28	5068	11	6.20-7.79	7.00 0.14	0.46	6.54
	Mount							
	Timpanogos	40.24	6000	8	7.00-7.68	7.38 -	-	-
	Provo	40.15	4549	18	6.10-7.62	7.10 0.08	0.36	5.01
	Vineyard	-	-	16	6.20-7.73	7.17 0.10	0.39	5.42
	Provo Canyon	-	5000	12	6.89-7.73	7.25 0.07	0.25	3.48
	Sevier Bridge							
	Reservoir	39.20	5000	11	6.29-7.71	7.04 0.13	0.43	6.07
	Piute							
	Reservoir	38.15	6000	10	6.30-7.43	6.93 0.11	0.34	4.92
	Parowan &							
	Cedar	37.50	5900	17	6.02-7.18	6.79 0.08	0.31	4.59
	Zion N. P.	37.20	4048	6	6.55-7.02	6.78 -	-	-
	Floy	38.56	4000	17	5.75-6.82	6.39 0.08	0.34	5.37
B	Arizona							
	Kayenta	36.44	6300	13	6.90-6.00	6.45 0.08	0.30	4.65
	Prescott	34.34	5280	133	6.00-7.90	7.01 0.03	0.32	4.56
	Phoenix	33.30	1092	11	6.55-7.28	6.85 0.07	0.22	3.21
	Globe	33.23	3541	22	6.50-7.60	7.03 0.06	0.27	3.84
C	Nevada							
	Gerlach &							
	Pyramid Lake	40.40	3900	10	6.40-7.32	6.90 0.11	0.35	5.10
	Reno & Verdi	39.32	4500	10	6.55-7.45	7.07 0.10	0.31	4.40
	Minden	38.58	4600	12	6.58-7.26	6.83 0.06	0.20	2.93
	Hawthorne &							
	Walker Lake	38.31	4326	27	6.57-7.95	7.26 0.07	0.36	5.01
	Caliente	37.36	4407	7	6.00-7.21	6.57 -	-	-

Width of Elytra				Diameter of Apical Dot			
Range mm	$\overline{X}$ ± SE	SD	CV	Range mm	$\overline{X}$ ± SE	SD	CV
2.45-2.81	2.67 0.03	0.11	3.97	0.42-0.73	0.59 0.02	0.09	14.51
2.50-2.90	2.69 0.02	0.10	3.61	0.35-0.83	0.63 0.03	0.14	21.75
2.52-2.89	2.75 -	-	-	0.50-0.71	0.62 -	-	-
2.36-3.01	2.67 0.02	0.14	5.17	0.45-0.83	0.63 0.01	0.08	13.14
2.52-2.93	2.71 0.01	0.10	3.46	0.34-0.76	0.63 0.01	0.09	14.11
2.30-2.83	2.62 0.03	0.14	5.42	0.40-0.75	0.55 0.02	0.09	15.73
2.49-2.82	2.65 0.02	0.09	3.25	0.39-0.80	0.65 0.02	0.08	12.91
2.30-2.90	2.70 0.02	0.12	4.44	0.43-0.80	0.63 0.01	0.09	14.83
2.34-3.10	2.72 0.02	0.14	5.29	0.28-0.84	0.63 0.01	0.11	16.83
2.40-2.96	2.73 0.03	0.12	4.47	0.50-0.73	0.64 0.02	0.08	12.75
2.49-2.85	2.70 0.02	0.11	4.19	0.41-0.90	0.64 0.02	0.10	15.02
2.31-2.86	2.64 0.04	0.15	5.72	0.52-0.73	0.64 0.02	0.06	9.89
2.54-2.75	2.65 0.02	0.07	2.56	0.44-0.77	0.63 0.02	0.09	14.60
2.31-2.87	2.62 0.03	0.14	5.46	0.40-0.89	0.64 0.02	0.11	16.72
2.42-2.95	2.71 0.02	0.12	4.46	0.40-0.83	0.67 0.02	0.09	14.09
				0.58-0.81	0.68 0.02	0.07	9.63
2.40-2.88	2.67 0.02	0.10	3.86	0.39-0.85	0.67 0.01	0.10	14.93
2.48-2.71	2.62 0.02	0.06	2.29	0.58-0.80	0.69 0.02	0.08	11.45
2.42-2.69	2.58 0.02	0.07	2.83	0.61-0.81	0.70 0.02	0.07	9.71
2.30-2.79	2.60 0.01	0.10	3.85	0.49-0.85	0.71 0.01	0.08	11.00
2.43-2.76	2.60 0.03	0.11	4.23	0.50-0.82	0.67 0.03	0.09	13.43
2.41-2.72	2.56 0.01	0.03	1.17	0.67-0.90	0.78 0.02	0.06	7.69
2.40-2.81	2.64 -	-	-	0.55-0.78	0.65 -	-	-
2.35-2.81	2.62 0.03	0.13	4.85	0.45-0.82	0.62 0.02	0.09	14.08
2.29-2.95	2.66 0.02	0.15	5.60	0.40-0.78	0.61 0.01	0.09	14.66
2.43-2.95	2.68 0.05	0.16	5.97	0.50-0.75	0.65 0.03	0.08	12.55
2.51-2.98	2.76 0.03	0.12	4.38	0.44-0.80	0.64 0.02	0.10	15.62
2.81-2.95	2.88 -	-	-	0.65-0.72	0.69 -	-	-
2.40-2.88	2.70 0.02	0.18	6.81	0.62-0.82	0.72 0.02	0.07	9.81
2.66-2.93	2.83 -	-	-	0.57-0.80	0.69 0.08	-	-
2.50-2.94	2.72 0.03	0.14	5.07	0.60-0.81	0.71 0.02	0.06	9.03
2.39-2.89	2.70 0.03	0.13	4.96	0.50-0.80	0.68 0.02	0.07	10.71
2.64-2.95	2.78 0.02	0.09	3.07	0.55-0.82	0.69 0.02	0.08	11.57
2.43-2.92	2.68 0.05	0.15	5.67	0.50-0.82	0.72 0.03	0.08	11.67
2.50-2.85	2.63 0.04	0.12	4.56	0.55-0.90	0.70 0.04	0.12	16.43
2.35-2.80	2.64 0.03	0.11	4.13	0.55-0.81	0.69 0.02	0.08	11.45
2.50-2.76	2.62 -	-	-	0.58-0.90	0.76 -	-	-
2.20-2.63	2.47 0.03	0.14	5.63	0.66-0.96	0.79 0.02	0.09	11.84
2.20-2.60	2.44 0.03	0.10	4.10	0.62-0.90	0.75 0.02	0.08	10.67
2.20-3.00	2.57 0.01	0.12	4.67	0.55-1.00	0.78 0.01	0.08	10.26
2.25-2.70	2.53 0.04	0.13	5.14	0.61-0.88	0.75 0.02	0.08	10.67
2.30-2.95	2.56 0.03	0.14	5.47	0.65-0.90	0.77 0.01	0.07	9.09
2.44-2.77	2.60 0.04	0.13	4.96	0.62-0.85	0.72 0.02	0.07	10.34
2.40-2.85	2.65 0.05	0.15	1.51	0.60-0.81	0.71 0.02	0.07	9.38
2.35-2.71	2.56 0.03	0.09	3.16	0.50-0.75	0.64 0.02	0.09	13.36
2.56-3.06	2.77 0.03	0.14	5.02	0.52-0.90	0.72 0.02	0.09	13.06
2.25-2.74	2.50 -	-	-	0.55-0.88	0.73 -	-	-

TABLE 5 - Variation in female *Cicindela oregona*.

						Length of Elytra		
Route	Locality	North lat.	Elev ft.	N	Range mm	$\overline{X}$ ± SE	SD	CV
A, B, C	Alaska							
	Tanana R.	63.00	1500	11	7.65-8.44	7.96±0.08	0.27	3.42
A, B, C	British Columbia							
	Terrace	54.31	223	16	6.56-8.61	7.86 0.12	0.48	6.04
	Q. Ch. Islands	53.00	0-4100	12	7.48-8.51	8.03 0.10	0.33	4.13
	Oliver	49.10	2143	24	6.85-8.57	7.79 0.10	0.48	6.21
A	Montana							
	Low. Medicine Lake	48.30	5000	21	6.95-8.25	7.69 0.08	0.37	4.81
	Hardy	47.11	3500	16	6.56-7.92	7.38 0.09	0.37	5.00
	Helena	46.35	4160	22	7.04-8.32	7.58 0.07	0.32	4.22
	Gardiner	45.03	5800	46	6.90-8.55	7.83 0.06	0.39	4.83
A	Wyoming							
	Yellowstone N.P.	44.30	7000	63	7.10-8.49	7.85 0.04	0.34	4.28
	Jackson Hole N.M.	43.50	6800	8	7.60-8.41	7.91 -	-	-
	Moran	43.52	6800	19	7.42-8.28	7.82 0.06	0.25	3.17
	11 miles S. Labarge	42.15	6600	5	7.55-7.91	7.75 -	-	-
	27 miles S. Labarge	42.14	6600	20	6.72-8.04	7.57 0.09	0.39	5.13
	Green River	41.33	6100	24	6.88-8.12	7.66 0.07	0.34	4.43
	Fort Bridger	41.19	6657	26	7.15-8.21	7.66 0.06	0.31	4.07
	Saratoga	41.28	6790	9	6.46-7.82	7.32 -	-	-
	Jelm	41.03	7500	42	6.80-8.48	7.57 0.06	0.36	4.73
A	Colorado							
	Estes Park	40.24	7547	13	6.98-7.90	7.43 0.08	0.30	3.97
	Trinidad	37.11	5999	10	6.89-7.79	7.23 0.09	0.29	4.05
A	New Mexico							
	Jemez Springs	35.45	6195	85	6.65-8.12	7.47 0.04	0.33	4.42
	Pecos & Beulah	35.34	7000	11	6.83-8.12	7.59 0.11	0.38	5.01
	Fort Wingate	35.30	6997	20	6.50-7.75	7.32 0.07	0.32	4.37
B, C	Idaho							
	Valley County	45.00	-	13	7.00-8.01	7.57 0.09	0.32	4.28
	Owyhee County	42.30	-	40	7.05-8.71	7.80 0.06	0.39	4.97
	Bear Lake	42.05	5924	58	6.45-8.37	7.62 0.06	0.45	5.92
B	Utah							
	Ogden	41.14	4296	3	7.32-7.73	7.59 -	-	-
	Salt Lake City	40.45	4354	21	6.72-8.40	7.71 0.11	0.49	6.41
	Alta	40.36	8585	11	7.34-8.32	7.90 0.09	0.30	3.84
	Stockton	40.28	5068	4	6.71-7.95	7.54 -	-	-
	Mount Timpanogos	40.24	6000	12	7.40-8.25	7.83 0.07	0.24	3.10
	Provo	40.15	4549	18	6.55-8.55	7.72 0.12	0.51	6.59
	Vineyard	-	-	10	7.08-8.31	7.82 0.12	0.38	4.87
	Provo Canyon	-	5000	14	7.10-8.32	7.83 0.08	0.31	3.96
	Sevier Bridge Reservoir	39.20	5000	10	7.12-7.87	7.52 0.09	0.28	3.71
	Piute Reservoir	38.15	6000	8	6.75-8.00	7.51 -	-	-
	Parowan & Cedar	37.50	5900	13	6.53-8.00	7.49 0.12	0.42	5.61
	Zion N.P.	37.20	4048	20	7.00-8.00	7.44 0.07	0.32	4.26
	Floy	38.56	4000	23	5.96-7.52	6.89 0.09	0.43	6.20
B	Arizona							
	Kayenta	36.44	6300	21	6.25-7.85	6.97 0.07	0.36	5.16
	Prescott	34.34	5280	133	6.70-8.50	7.65 0.03	0.38	4.96
	Phoenix	33.30	1092	15	7.10-8.10	7.54 0.09	0.33	4.38
	Globe	33.23	3541	34	6.62-8.10	7.56 0.07	0.40	5.33
C	Nevada							
	Gerlach & Pyramid Lake	40.40	3900	11	7.32-8.20	7.66 0.10	0.33	4.35
	Reno & Verdi	39.32	4500	10	6.24-8.22	7.58 0.21	0.67	8.84
	Minden	38.58	4600	12	7.57-8.10	7.82 0.05	0.18	2.24
	Hawthorne & Walker Lake	38.31	4326	27	6.46-8.60	7.88 0.05	0.49	6.19
	Caliente	37.36	4407	6	6.80-7.72	7.13 -	-	-

Width of Elytra				Diameter of Apical Dot			
Range mm	$\overline{X}$ ± SE	SD	CV	Range mm	$\overline{X}$ ± SE	SD	CV
2.93-3.20	3.08±0.03	0.08	2.71	0.58-0.91	0.73±0.03	0.09	12.98
2.50-3.33	3.07 0.05	0.19	6.22	0.45-0.90	0.72 0.03	0.13	18.33
2.85-3.22	3.08 0.03	0.12	3.90	0.62-0.88	0.76 0.02	0.08	10.50
2.73-3.29	3.07 0.04	0.18	5.93	0.59-0.91	0.74 0.02	0.09	12.59
2.59-3.28	3.02 0.04	0.20	6.49	0.42-0.82	0.68 0.03	0.12	17.21
2.59-3.21	2.93 0.04	0.16	5.43	0.58-0.80	0.68 0.02	0.07	10.04
2.69-3.29	2.98 0.04	0.17	5.57	0.57-0.90	0.72 0.02	0.10	13.89
2.69-3.76	3.09 0.03	0.19	6.18	0.38-0.91	0.74 0.01	0.10	13.70
2.81-3.34	3.10 0.02	0.13	4.32	0.50-0.92	0.76 0.01	0.10	12.62
3.30-3.35	3.16 -	-	-	0.69-0.99	0.81 -	-	-
2.85-3.20	3.07 0.02	0.10	3.26	0.61-0.88	0.75 0.02	0.08	10.43
2.91-3.05	3.02 -	-	-	0.68-0.85	0.74 -	-	-
2.66-3.10	2.95 0.03	0.13	4.34	0.54-0.89	0.71 0.02	0.11	14.79
2.63-3.25	3.01 0.03	0.15	4.95	0.51-0.93	0.74 0.02	0.09	12.59
2.72-3.24	3.02 0.02	0.13	4.14	0.64-1.00	0.80 0.02	0.08	10.60
2.49-3.05	2.84 -	-	-	0.63-0.92	0.78 -	-	-
2.70-3.30	3.00 0.02	0.14	4.53	0.60-0.94	0.78 0.01	0.07	9.18
2.63-3.17	2.88 0.04	0.15	5.10	0.68-0.86	0.77 0.01	0.05	6.49
2.68-3.04	2.86 0.04	0.13	4.51	0.70-0.91	0.81 0.02	0.07	8.27
2.60-3.20	2.94 0.01	0.13	4.42	0.72-1.05	0.87 0.01	0.08	8.25
2.70-3.24	3.00 0.05	0.15	5.00	0.75-1.00	0.88 0.02	0.08	9.09
2.56-3.17	2.95 0.03	0.13	4.41	0.80-1.04	0.92 0.02	0.08	8.70
2.75-3.20	2.98 0.04	0.15	5.03	0.60-0.83	0.72 0.02	0.06	8.00
2.68-3.41	3.07 0.02	0.15	4.66	0.55-1.00	0.77 0.01	0.09	12.12
2.52-3.40	3.01 0.02	0.18	5.87	0.38-1.00	0.74 0.01	0.11	15.41
3.00-3.02	3.01 -	-	-	0.70-0.80	0.73 -	-	-
2.78-3.38	3.08 0.04	0.18	5.97	0.65-0.96	0.79 0.02	0.08	10.62
2.91-3.30	3.14 0.04	0.12	3.76	0.70-1.02	0.80 0.03	0.09	11.19
2.89-3.20	3.09 -	-	-	0.72-0.96	0.88 -	-	-
2.96-3.25	3.13 0.02	0.08	2.55	0.60-0.95	0.83 0.03	0.10	12.53
2.54-3.39	3.07 0.05	0.21	6.91	0.70-0.98	0.86 0.02	0.08	9.77
2.82-3.32	3.09 0.05	0.16	5.28	0.70-0.91	0.81 0.03	0.09	10.90
2.86-3.31	3.09 0.04	0.13	4.30	0.57-0.89	0.76 0.03	0.10	13.16
2.80-3.18	2.95 0.03	0.11	3.76	0.76-0.95	0.84 0.02	0.06	6.87
2.63-3.21	2.95 -	-	-	0.70-0.99	0.84 -	-	-
2.79-3.17	3.00 0.03	0.12	3.83	0.80-0.97	0.86 0.02	0.06	7.50
2.85-3.19	3.01 0.02	0.09	3.14	0.74-1.00	0.87 0.02	0.07	8.33
2.43-3.05	2.77 0.04	0.17	6.14	0.74-1.05	0.91 0.02	0.08	8.77
2.30-2.90	2.66 0.03	0.16	5.83	0.75-1.10	0.89 0.02	0.10	11.24
2.50-3.30	2.93 0.01	0.16	5.51	0.75-1.20	0.93 0.01	0.09	9.68
2.60-3.05	2.84 0.04	0.14	4.93	0.75-1.05	0.90 0.02	0.08	8.89
2.40-3.20	2.85 0.03	0.18	6.32	0.65-1.05	0.90 0.01	0.08	8.89
2.76-3.33	3.04 0.05	0.16	5.30	0.60-0.94	0.78 0.03	0.09	11.47
2.45-3.13	2.94 0.07	0.24	8.06	0.65-0.87	0.79 0.02	0.07	9.48
2.95-3.16	3.06 0.02	0.08	2.61	0.61-0.98	0.79 0.03	0.10	12.03
2.50-3.35	3.10 0.04	0.20	6.39	0.60-1.00	0.84 0.02	0.10	11.67
2.70-2.89	2.82 -	-	-	0.70-0.92	0.81 -	-	-

and Yellowstone National Park, Wyoming in table 5.

Populations in Nevada are generally longer and wider than samples from Utah, Colorado, and Wyoming at similar elevations; but east-west clines are very irregular.

Specimens collected in Queen Charlotte Islands, British Columbia are the only insular members recorded in tables 4 and 5. The *oregona* females of these Islands have a higher mean value for length of elytra than have those from any other locality listed in table 5, and the mean value for males is slightly less than those of Alta and Mount Timpanogos, Utah. However, the male samples from the Queen Charlotte Islands, Mount Timpanogos, and Alta each are represented by fewer than 10 specimens and a more accurate comparison can be made with the females. Individuals from localities of high elevation in Utah, and Wyoming are scarcely broader than are those of Queen Charlotte Islands (see table 5).

A relationship of body size with latitude or altitude is evident in many animal species other than *Cicindela oregona*. North American brown bears for example, increase in size as the latitude increases (Rausch 1963). This phenomonon has also been shown in many species of birds (Mayr 1942, 1963, Hamilton 1961). In insects, honey bees and two species of European *Carabus* vary in the same way (Mayr 1963, p. 326). New Guinea dragon flies have been found to increase in size at higher elevations also (Mayr 1963, p. 326). In ectothermal animals as a whole the largest body size may just as often be found in the warmest portion of a species range. Lindroth (1963) noted that in some Carabids of Newfoundland dwarf forms are frequently confined to high altitudes or marginal northern areas of the species range. Ball (1959) observed that several species of the ground beetle genus *Dicaelus* were larger in southern areas of their ranges. Likewise the small, flightless grasshopper *Melanoplus puer* shows a general southward increase in size (Hubbell 1956). Mayr (1963) presents a review of evidence for the adaptive nature of geographic variation in which latitudinal and altitudinal changes are discussed in relation to geographic variation in body size.

Color pattern of the elytra - While body size of *Cicindela oregona* decreases from north to south the breadth of the white markings of the elytra increases. The diameter of the apical dot increases approximately 0.007 mm for each degree of latitude. Data on the expanse of elytral pattern, illustrated by the diameter of the apical dot, are presented in table 4 for males and in table 5 for females.

In the first route (A) from Alaska to New Mexico there is a slight uniform increase in apical dot size through British Columbia and Montana. In Fort Bridger, Wyoming the cline is steeper and continues to increase through Colorado to New Mexico. Fort Wingate, New Mexico is represented by individuals having a very wide apical dot with an average measurement of 0.78 mm. The difference between the mean values for males from Fort Wingate and those from Pecons, New Mexico is statistically significant, but this is not so for females. The same applies to material from Hardy, Montana and from Helena and Lower Medicine Lake, Montana.

A similar latitudinal increase in the diameter of the apical dot is evident in course (B) from Alaska to Arizona. There is a slight decrease in the mean apical dot size of males in southern Idaho (this is not true for females) but the color pattern expands markedly in Utah. Populations with the widest apical dots are present in Floy, Utah, and Prescott and Globe, Arizona. A similar cline exists in the third path of population samples that extends from Alaska to Nevada.

Coloration - The elytra, thoracic pleura, pronotum and, indeed, the entire body are subject to color variation in this species. The elytra may be brown, green, purple, blue and occasionally very dark brown that is almost black. The thoracic pleura are coppery, metallic blue, purple or green. Color of the pronotum and elytra is generally the same on each individual. Unicolored specimens occur throughout the species range in scattered localities and they are usually green, less frequently blue. In Arizona, southern Nevada, and southern New Mexico a green pronotum is usually associated with purple elytra. North of the limits of populations that have purple elytra the beetles are usually metallic blue-green ventrally and brown dorsally. Figure 19 is a pie-graph map illustrating color variation of the elytra and thoracic pleura in populations in the northern portion of the range of *oregona*. Each "pie" represents a single population sample. Figure 20 is a southwestern continuation of figure 19.

Coppery thoracic pleura are prevalent throughout northwestern British Columbia, Yukon Territory and Alaska. In figure 19 samples A (Tanana River, Alaska) to K (Helena, Montana) coppery thoracic pleura are most frequent, followed in numbers by metallic green, blue and purple, in that order. In Alaska, the Yukon Territory and northern British Columbia, only the coppery condition exists. In samples D, E, F and G, in central British Columbia, individuals with metallic blue-green or purple thoracic pleura are present in low frequency. Southward, coppery thoracic pleura are prevalent in central British Columbia, on the eastern slopes of the Rocky Mountains in Alberta and Montana, and in eastern Idaho, Utah, Wyoming, Colorado, northeastern Arizona and northern New Mexico (see data for Fort Bridger, Wyoming and Jemez Springs, New Mexico in pictorialized scatter diagrams figures 25 and 27). In British Columbia (fig. 19) coppery thoracic pleura are abruptly replaced by metallic purple thoracic pleura and this condition extends throughout southern British Columbia from the Pacific coast east to the Continental Divide. Of the individuals represented in samples L to U, only eight have coppery thoracic pleura in this region. Specimens with metallic green thoracic pleura are common in coastal populations of British Columbia and Washington, and also along the eastern ridge of the Rocky Mountains. This condition is less common in Oregon and California where metallic purple and metallic blue thoracic pleura are dominant (fig. 20). Thoracic pleura of specimens of Owyhee County, Idaho (L) are predominantly metallic purple, also. Populations with thoracic pleura ranging from coppery to metallic purple through metallic blue and metallic green are found in northern Utah near Alta and southern Utah in the area of Zion National Park. Across southwestern United States from San

Diego, California to Mountain Park, New Mexico, specimens with metallic purple thoracic pleura are most frequent.

Brown elytra are most common throughout most of the range of *oregona*. Brown color is entirely replaced by purple color in central Arizona, southern New Mexico and southern Nevada. This situation is discussed in detail in the subspecies section. Populations that are highly variable in color of elytra occur throughout the range of this species. Such populations are on the Pacific coast from Alaska to southern California and almost all individuals with blue or green elytra exist in coastal localities (see figs 19, 20). Brown is most common, followed by green, and then blue. In Garibaldi Park, British Columbia, a very large proportion of specimens with blue elytra are present, while Vancouver and Victoria, British Columbia populations are made up mainly of individuals with green elytra. In Humbolt County, California and Port Orford, Oregon, most of the specimens have brown elytra, and some members with green and blue elytra are also present. Likewise, a few individuals with blue and green elytra are present in San Francisco and in San Diego, California. Variation in elytral color is the rule in northern and southern Utah, where blue, green, and brown specimens are present.

Shelford (1914) studied color and color pattern of tiger beetles and he found, of the species studied in detail, the more brilliant colors occur in warm, arid localities, and extended markings in cooler regions. These findings apply only in part to *oregona*. In Arizona, New Mexico and southern Nevada, which are warm dry areas, specimens with bright metallic dorsal surfaces are prevalent but brilliant blue and green specimens of *oregona* also live along the Pacific coast from California to Alaska, and this is quite a humid zone. The markings of *oregona* are expanded in warmer localities and this condition contrasts with the results obtained by Shelford regarding pattern of elytra.

The pattern of variation - Independent character changes have resulted in discordant variation within *C. oregona*. Elytra are dark brown, generally, in the northern, eastern and western areas of the range but they are purple in the south, and very light brown in eastern Utah. In contrast, pleura usually blue to purple in the west and south, are coppery in northern and eastern portions of the range. In addition general body size decreases clinally from the north to south and also varies from higher to lower altitudes. Finally extent of white markings on elytra increases southward. Although recognition of subspecies in species that show discordant variation is controversial, (p. 90 , and Inger 1961) I think it is useful to group into subspecies the population samples of *oregona*.

Maintenance of variation in this species appears to be largely dependent on geographical factors but may also be due to variation in the season of occurrence of adults. Mature specimens of my own and other collectors from boreal populations appear to be most plentiful for June, July, and August (based on specimen label information and personal collecting). This seems to be true for alpine populations in southern regions as well. Adult specimens from populations in desert areas of Utah, Arizona, and New Mexico have been collected from March to October inclusive. This suggests that they are common throughout this time but

Fig. 19. Pie-graph map illustrating geographic variation in the color of the elytra and thoracic pleura of some populations samples of *Cicindela oregona* . The numbers of specimens with a given color combination are indicated opposite the appropriate section. Thus, 20 placed at 1 o'clock signifies that 20 specimens have coppery thoracic pleura and brown elytra.

Elytra

BLUE · BROWN

Pleura

GREEN

OREGON

California:
a – San Francisco   h – Santa Cruz Island
b – Antioch   i – Los Angeles
c – Yosemite National Park j – Colton
d – Bigpine   k – Victorville
e – Lone Pine and Olancha l – San Jacinto Mountains
f – Coalinga   m – San Diego
g – Carmel   n – Plumas County

OREGON:
A – Portland
B – Hood River and Dalles
C – Umatilla
D – Florence
E – Charleston
F – Port Oxford and
    Humbug Mt.
G – Medford
H – Eugene
I – Lake Albert
J – Steens Mountain
K – Malheur Lake
IDAHO:
L – Owyhee County
CALIFORNIA:
M – Gottville
N – Dillon Creek
O – Eureka and Trinidad
P – Hoopa Valley
Q – Douglas
R – Bridgeville and Mad
    River
S – Miranda and Fort
    Seward
T – Lassen National Pk.
U – Lake Tahoe
NEVADA:
V – Walker Lake
CALIFORNIA:
W – Bucks Lake
X – Chico
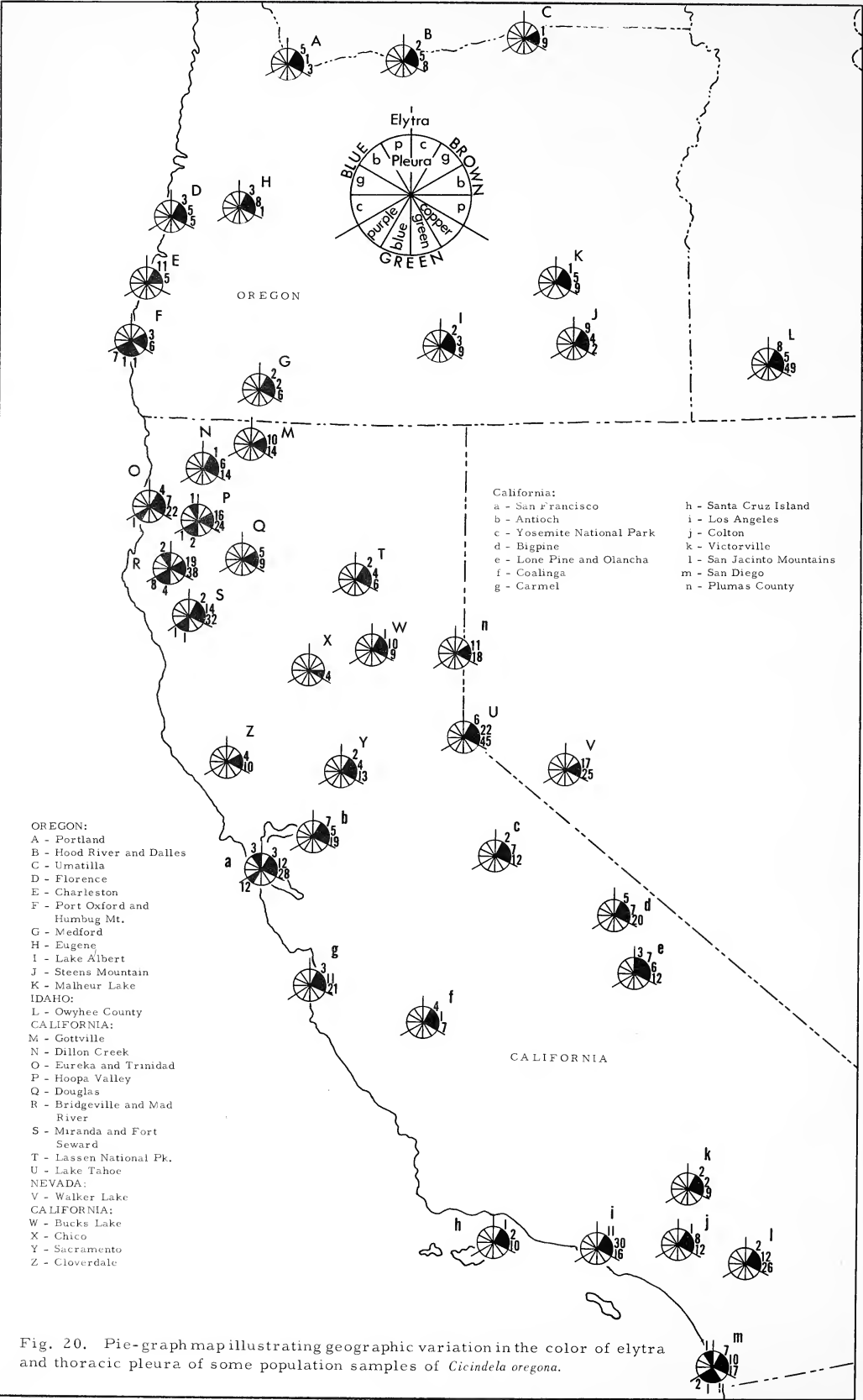Y – Sacramento
Z – Cloverdale

CALIFORNIA

Fig. 20. Pie-graph map illustrating geographic variation in the color of elytra and thoracic pleura of some population samples of *Cicindela oregona*.

I do not believe so. Because *C. oregona* is riparian, size of populations, in arid southwestern regions, is likely to fluctuate with rain. Kendrew (1961) points out that rainfall is variable in these desert regions with a maximum in late summer and winter, and that the mountain ranges which rise on the southwestern plateau have rather more rain. It follows that activity peaks of desert populations probably do not occur at the same time year after year but in different periods in relation to rainfall. Attempts to collect desert forms during winter months have been unsuccessful. Perhaps they are most numerous during late summer-later than the peak of alpine populations.   Such asynchronous number fluctuations effect a reduction in gene flow and thus maintain the variation between desert and alpine populations of the southwest.

Subspecies - I recognize four subspecies of *Cicindela oregona*.   I have followed the 75% rule in defining the group taxonomically (see p. 90 , and Mayr *et al.* 1953). The differences between two or more populations in two or more characters are best illustrated by a pictorialized scatter diagram. Ten such diagrams and a locality map of the population samples compared in the scatter diagrams are presented as figs 21-31. Subspecies can be readily distinguished from each other on the basis of one or more external characters.   Males and females are treated separately.   Generally five localities are represented in each diagram.   Each locality is represented by ten specimens or less, and they have been selected randomly. Fifty specimen symbols are placed on a diagram.

1	Thoracic pleura blue or purple...........................	2
	Thoracic pleura coppery.................................	3
2	Elytra purple, elytral pattern broad, pronotum green....... .... *o. maricopa*	
	Elytra brown, green, blue, or rarely purple; elytral pattern narrow; pronotum brown.................... *o. oregona*	
3	Elytra light brown; elytral pattern broad........... *o.. navajoensis*	
	Elytra dark brown; elytral pattern narrow........... *o. guttifera*	

The nominate subspecies *Cicindela oregona oregona* ranges from southern British Columbia in Canada,   to southwestern California. it is present throughout Washington, Oregon, and California, except for the southeast portion of that state.   The Continental Divide serves as the eastern limit in the north, from Banff, Alberta south to Yellowstone National Park,   Wyoming.   Further south *oregona oregona* is found as far east as Owyhee Count, Idaho, western Nevada and finally near the southern portions of the Sierra Nevada Mountains in California (fig. 18). A combination of green, blue, or brown elytra with metallic purple or blue thoracic pleura is characteristic of this subspecies. Individuals with green elytra are numerous in or near the above localities.   The scattered occurrence of these blue and green individuals along the Pacific coast may be evidence of a blue form that was once widespread in these

coastal regions, but was infiltrated by a more vigorous stock, character-
ized by the possession of brown elytra. Whenever these two aggregates
of populations came into contact introgression took place and the presence
of green individuals interpreted as hybrids, marks what once were zones
of contact. In more southern locations these green and blue forms have
been all but completely replaced by brown. On the other hand they may
be recent phenotypes whose gene complex originated in southwestern
British Columbia when the blue phenotypes were relatively common.
Another possibility is that the green and blue forms might be ecopheno-
types. Although Shelford observed that *Cicindela tranquebarica* Herbst is
green on the coasts and coastal mountains and also that in *Cicindela scutellaris*
green forms were most common along the Atlantic coast (Shelford 1917),
he did not believe that this was the result of direct influence of the en-
vironment on the phenotypes.

     The subspecies *o. oregona* comes in contact with *o. guttifera* LeConte
in southern British Columbia and along the slopes of the Rocky Mountains
from Banff, Alberta to Yellowstone National Park, Wyoming. Many
specimens that appear to be hybrids are present in areas of contact of
these two subspecies, and such are distinguished by their metallic
green thoracic pleura (fig. 19).

     In San Diego, California a hightly variable group of populations
is present (fig. 20), for in this area specimens typical of both *o. oregona*
and *o. maricopa* occur. This situation could be the result of *maricopa* genes
infiltrating the more numerous *oregona* population in the region (figs 22,
23). Only five phenotypically *maricopa* specimens are known from the San
Diego area.

     Cicindela o. guttifera ranges the Rockies from Fort Yukon, Alaska to
northern New Mexico (fig. 18). In Alaska and north and central British
Columbia, *guttifera* ranges from the Pacific coast to the eastern slopes of
the Rockies but continues southward in a very narrow zone to northern
New Mexico. This subspecies also occurs in northern and central Utah.
Coppery thoracic pleura and brown elytra that have a metallic lustre
characterize it. I have already mentioned that hybridization takes place
between *oregona guttifera* and *oregona oregona* in much of eastern Idaho and
western Montana, and intermediate specimens with metallic green sides
are not uncommon in northern and central Utah where they are distributed
through *oregona guttifera* populations. In southwestern Utah a highly variable
series of populations occurs, consisting of individuals ranging from typ-
ical *guttifera* to typical *maricopa* . This region is undoubtedly a melting pot
of these two subspecies (figs. 24, 25). It may be argued that this var-
iation is a result of hybridization between *oregona* and *maricopa* and not as
above. This is not likely since at the present time *oregona* is uncommon in
eastern Nevada and it is not abundant in Utah, but it may have contributed
to this variation in pluvial times.

     The ranges of *Cicindela o. navajoensis* and *guttifera* come very close in
northwestern New Mexico (fig. 18). *Navajoensis* is relatively small in size
and has much lighter brown elytra and broader pattern of elytra than
guttifera. Like the latter, *navajoensis* has coppery thoracic pleura. A
color character gradient occurs from Kayenta, Arizona to Jemez Springs,
New Mexico through an intermediate locality, Fort Wingate, New Mexico.

Color of the elytra in Kayenta is light brown, dark brown in Jemez
Springs and intermediate in Fort Wingate. Fort Wingate specimens are
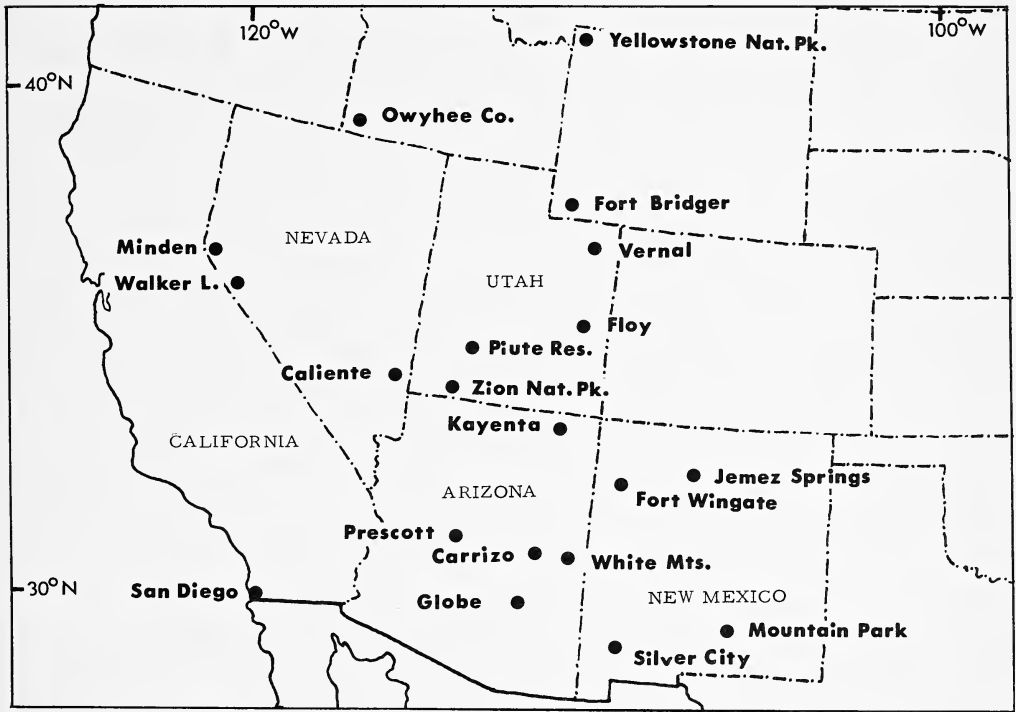also intermediate in lengths and widths of elytra and the apical dot (figs
26, 27).

Fig.21 - Locality map of population samples of *C. oregona* LeConte compared in scatter diagrams.

The geographic ranges of *oregona* and *navajoensis* are not in contact;
their morphological relationships are demonstrated in figs 28 and 29.
In southeastern Arizona and southwestern New Mexico are variable
populations that consist of individuals structurally between *navajoensis* and
maricopa (figs 30, 31). The members of these groups are generally smaller
in body size than *maricopa* and larger than *navajoensis*. Their elytra are
mainly purplish brown. The Fort Wingate sample in figures 30 and 31 is
not pure *navajoensis* but is intermediate between *navajoensis* and *guttifera* (figs
26, 27). Thus the specimens appear as intermediates between *navajoensis*
and *maricopa* in figures 30 and 31.
Specimens of *o. maricopa* have brilliant purple elytra, brown to met-
allic green pronota, and metallic purple thoracic pleura. *Maricopa* is dis-
tributed sparsely through southern California, southeastern Nevada, and
southern New Mexico, but it is common in central and southern Arizona
(fig. 18). California "*maricopa*" may be a minor element in predominantly
o. oregona populations and if so they are *maricopa* in a typological sense only.
The form of this and other subspecies of *Cicindela oregona* has been compared

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Figs. 22 to 31. Pictorialized scatter diagrams illustrating character differences between population samples of *C. oregona oregona* (⊚), *C.o. maricopa* (◯), *C.o. guttifera* (●), and *C.o. navajoensis* (◌). Intermediate populations represented by divided circles (⦶ ◑ ◐ ⊘); elytral color by vertical bars: long - purple, medium - green, short - blue, no bar - brown; pleural color by horizontal bars: long - purple, medium - green, short - blue, no bar - coppery. Males above, females below.

Fig. 24 ♂

Fort Bridger, Wyoming
Piute Reservoir, Utah

Caliente, Nevada
Prescott, Arizona

Zion Nat. Pk., Utah

WIDTH OF ELYTRA / DIAMETER OF APICAL DOT

5.00

3.00

6.50   7.50

LENGTH   OF   ELYTRA   MM

Fig. 25 ♀

WIDTH OF ELYTRA / DIAMETER OF APICAL DOT

4.00

3.00

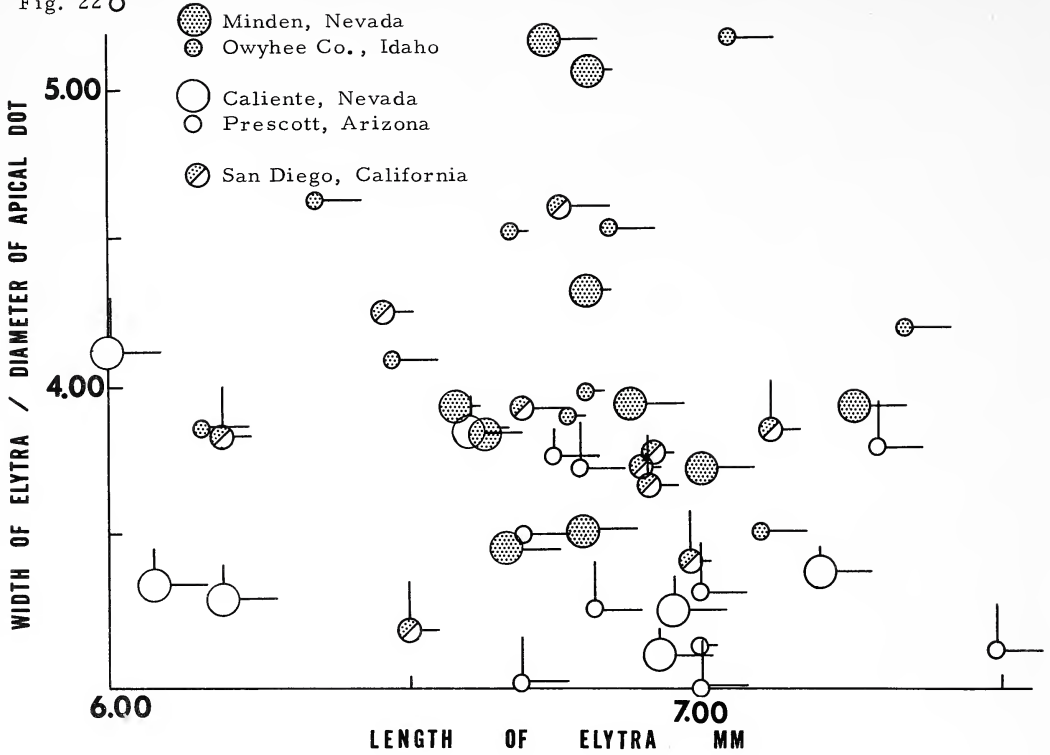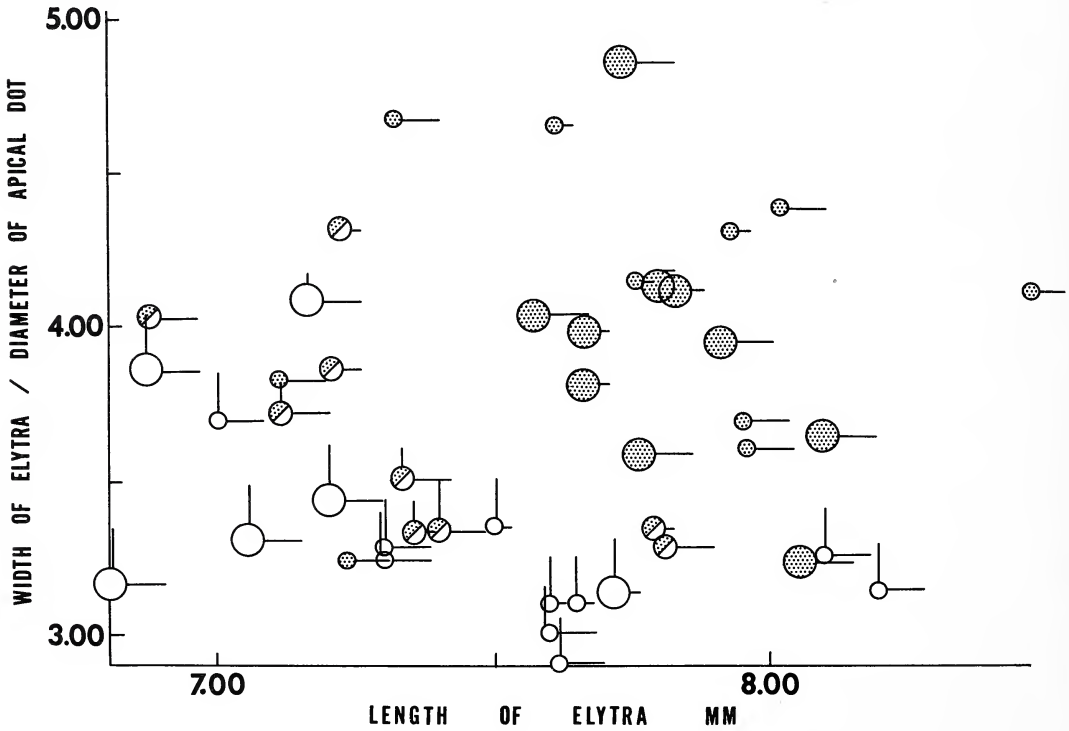7.00   8.00

LENGTH   OF   ELYTRA   MM

Fig. 26 ♂

Fig. 27 ♀

Figs. 22 to 31. Pictorialized scatter diagrams illustrating character differences between population samples of *C. oregona oregona* (◐), *C.o. maricopa* (◯), *C.o. guttifera* (●), and *C.o. navajoensis* (◑). Intermediate populations represented by divided circles (◐ ◐ ◑ ◑); elytral color by vertical bars: long - purple, medium - green, short - blue, no bar - brown; pleural color by horizontal bars: long - purple, medium - green, short - blue, no bar - coppery. Males above, females below.

Fig. 28 ♂
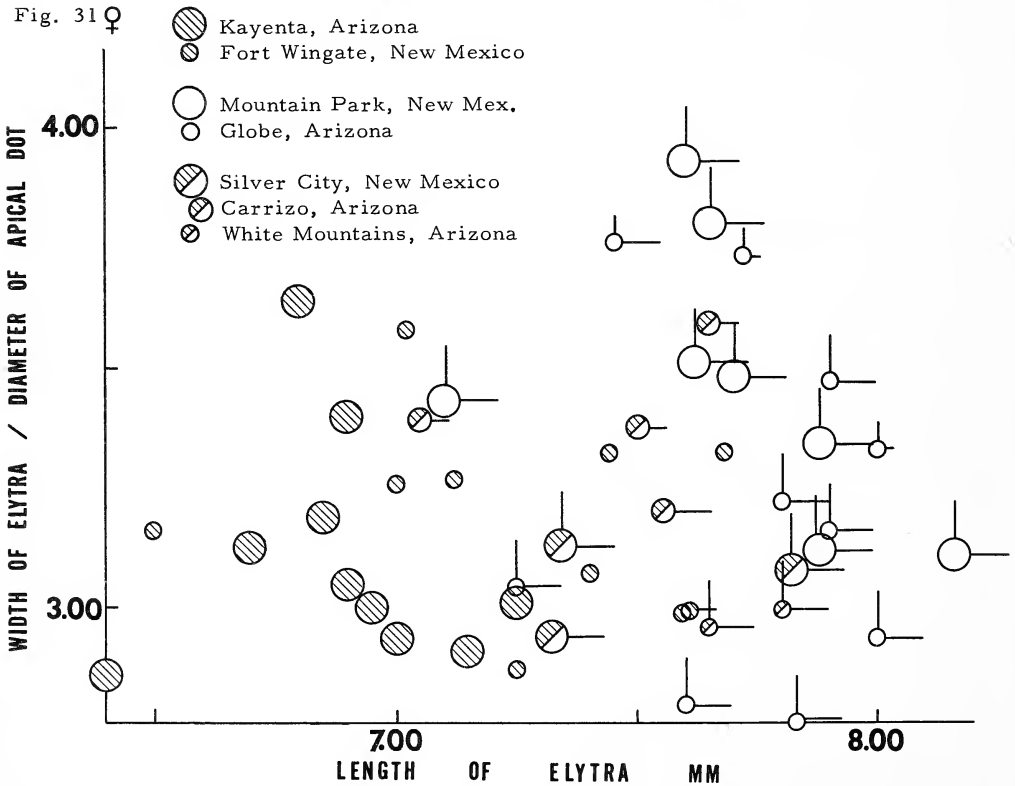
Yellowstone Nat. Pk.,
        Wyoming
Walker Lake, Nevada
Owyhee Co., Idaho

Kayenta, Arizona
Floy, Utah

WIDTH OF ELYTRA / DIAMETER OF APICAL DOT

5.00

3.00

6.00   7.00   8.00

LENGTH   OF   ELYTRA   MM

Fig. 29 ♀

WIDTH OF ELYTRA / DIAMETER OF APICAL DOT

5.00

3.00

7.00   800

LENGTH   OF   ELYTRA   MM

132

Fig. 30 ♂

Fig. 31 ♀

⊘ Kayenta, Arizona
⊘ Fort Wingate, New Mexico

◯ Mountain Park, New Mex.
○ Globe, Arizona

⊘ Silver City, New Mexico
⊘ Carrizo, Arizona
⊘ White Mountains, Arizona

Figs. 22 to 31. Pictorialized scatter diagrams illustrating character differences between population samples of *C. oregona oregona* (⊘), *C.o. maricopa* (◯), *C.o. guttifera* (●), and *C.o. navajoensis* (⊗). Intermediate populations represented by divided circles (⊘ ● ⊗ ⊘); elytral color by vertical bars: long - purple, medium - green, short - blue, no bar - brown; pleural color by horizontal bars: long- purple, medium - green, short - blue, no bar - coppery. Males above, females below.
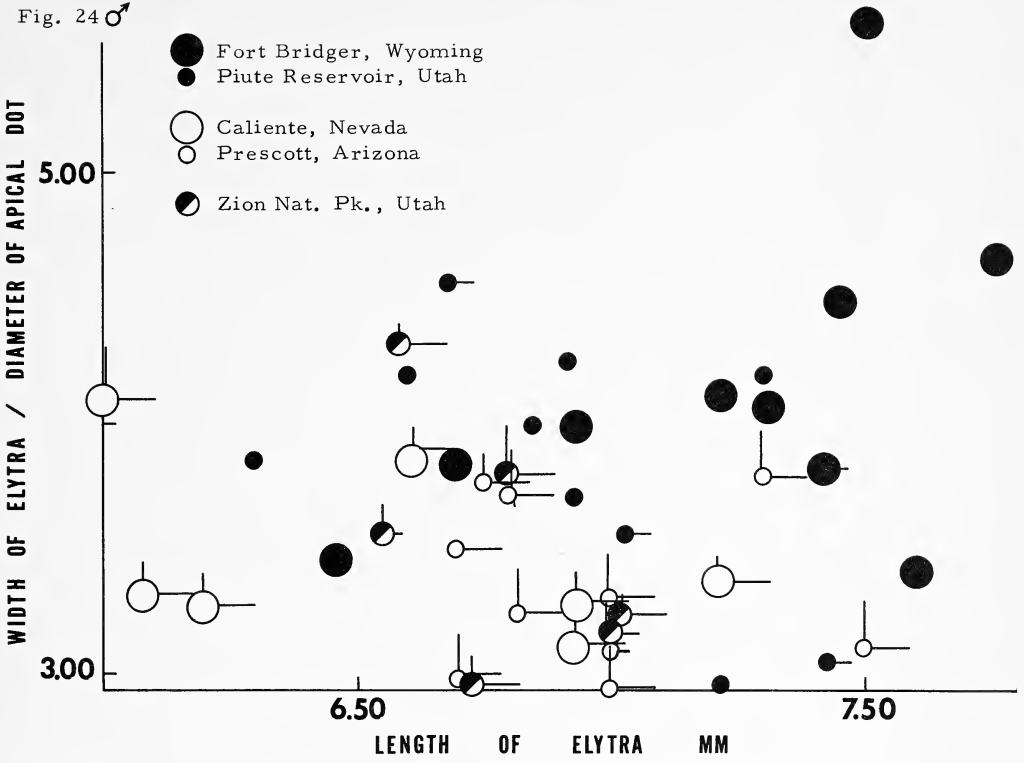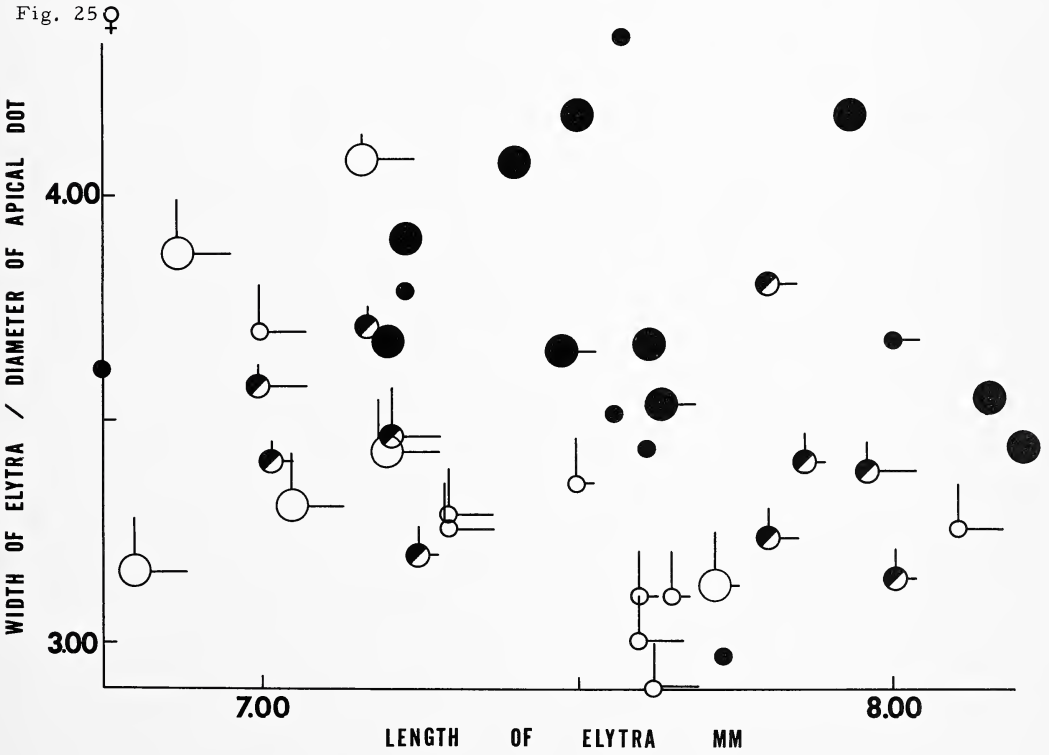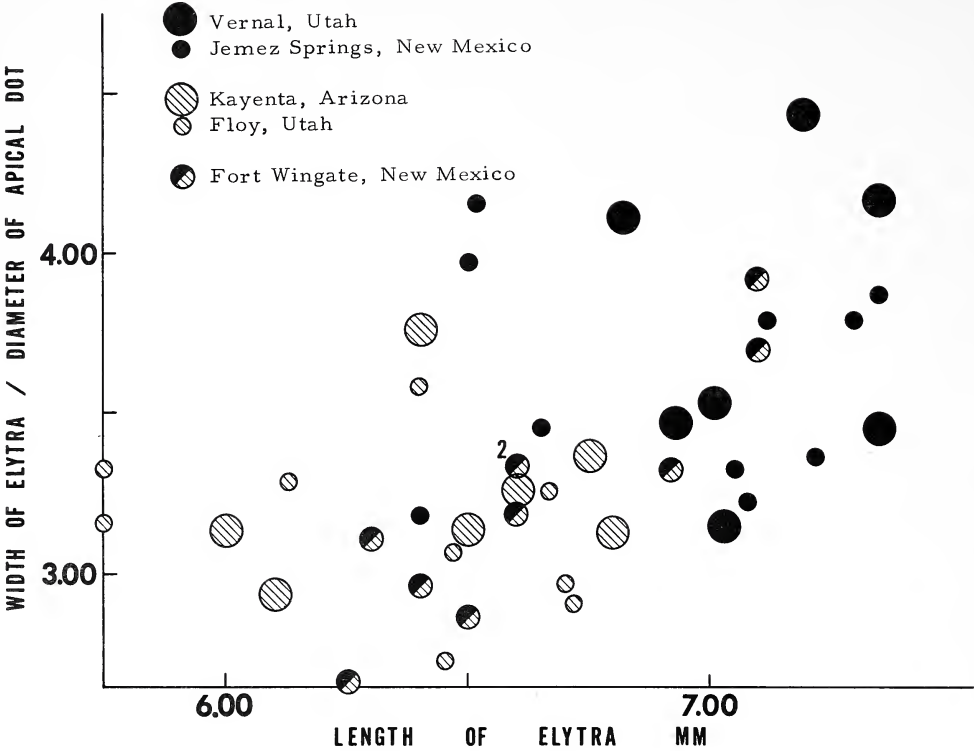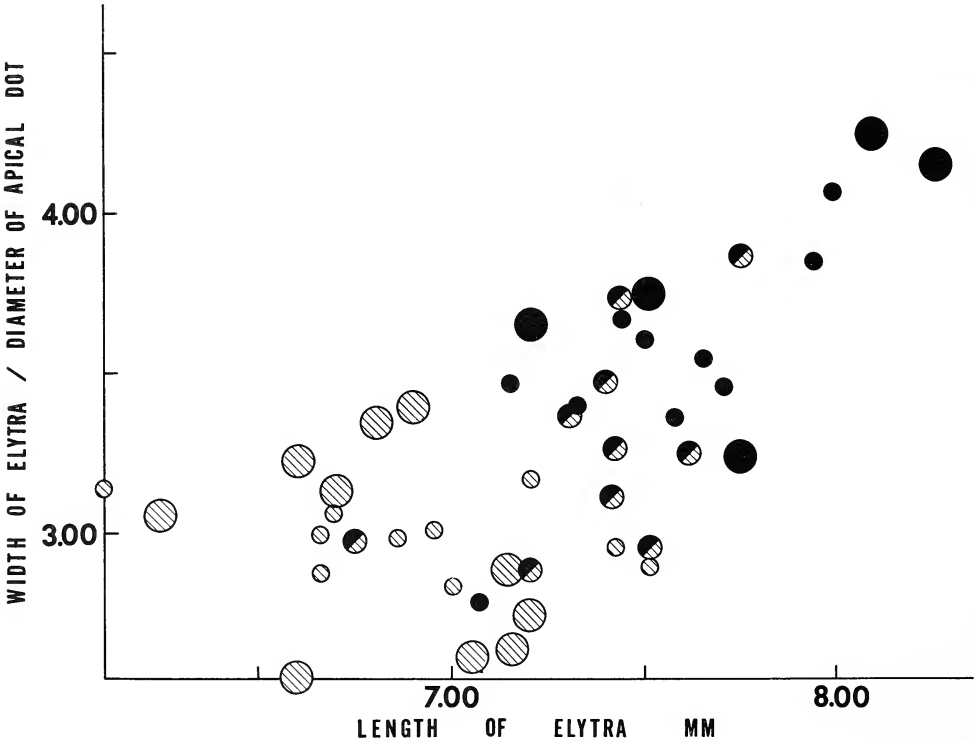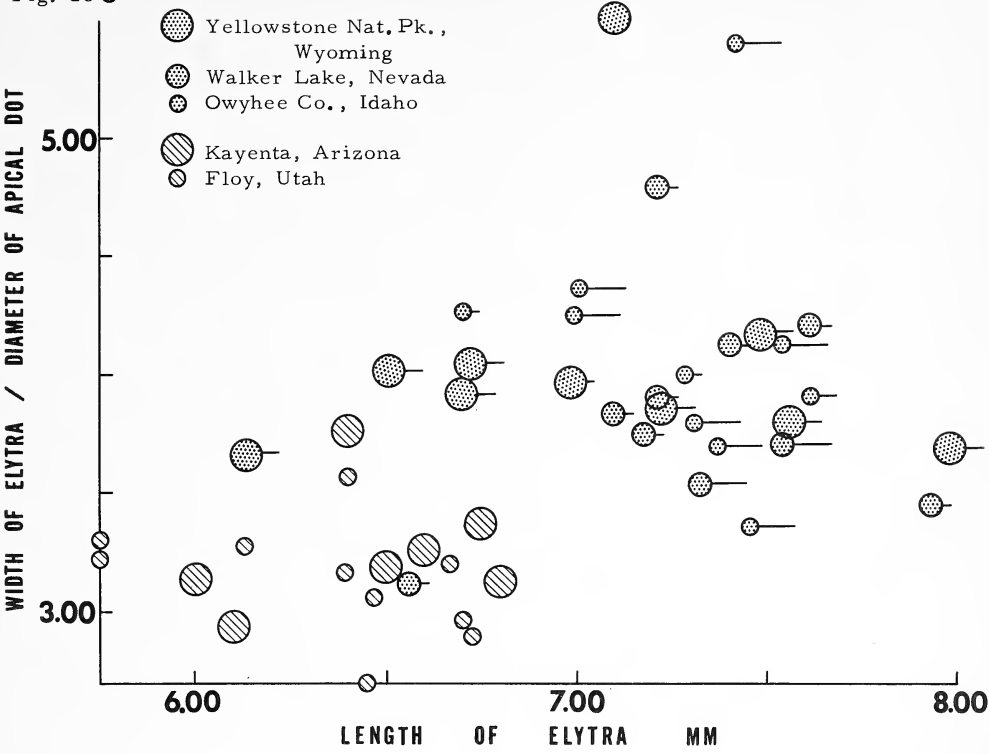
above.

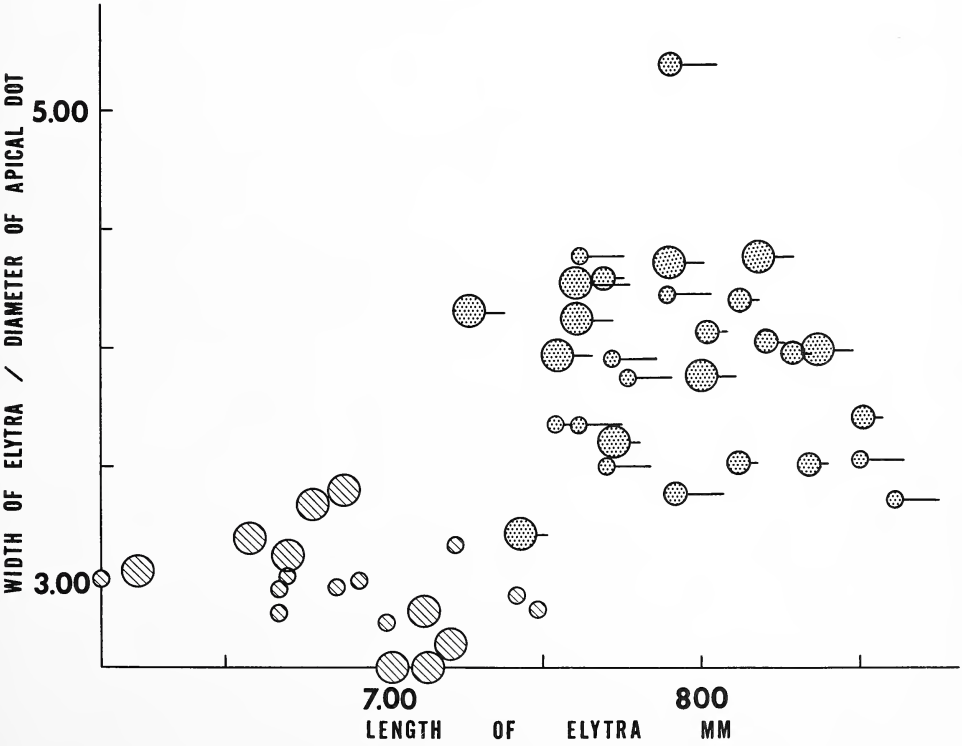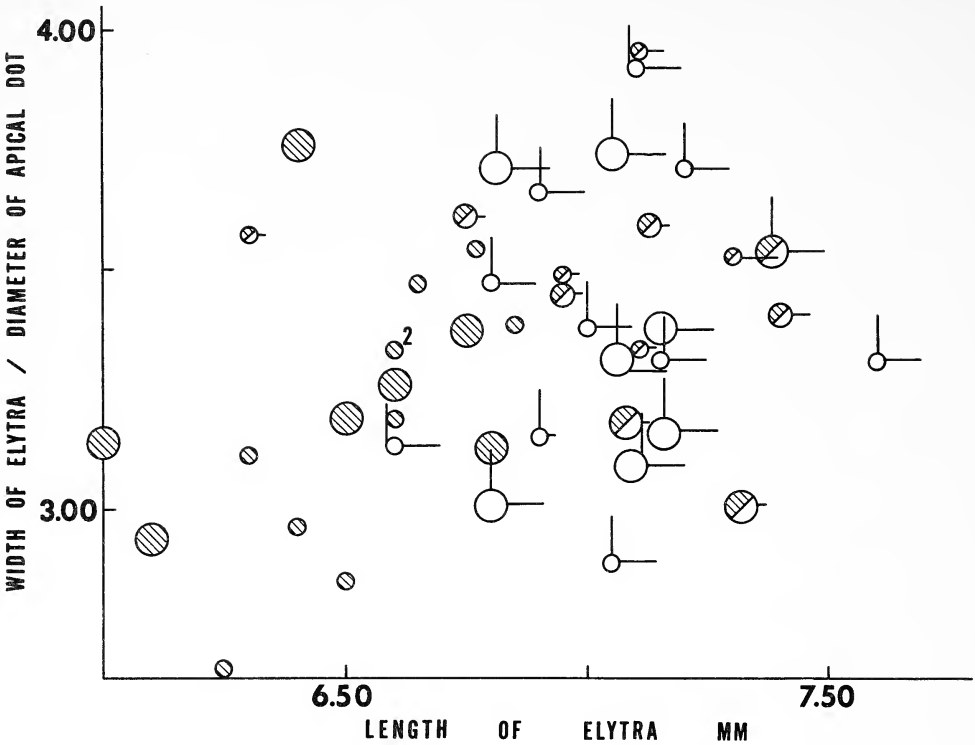*History of Distribution and Subspeciation*

Distribution of *C. oregona* is restricted to western United States and Canada bounded by Alaska, southern California, Arizona and New Mexico and the Rocky Mountains. Within this area four geographically distinct groups of populations exhibit boundaries that are generally barriers such as deserts and mountain ranges (fig. 18). All population samples of these subspecies that I have examined, have been collected in the above described range of *C. oregona*. I have seen two *maricopa* specimens, however, that are labelled "Texas", but specific localities are not given. They could have been collected in western Texas near *maricopa* localities in New Mexico. Because the total range of *C. oregona* is well marked and no populations occur in remote regions outside of the described range it appears highly likely that subspeciation took place somewhere in western North America.

Before discussing further the questions of how and when formation of subspecies occurred in *oregona* it should be emphasized that determination of evolution of a subspecies without a fossil record is a highly speculative matter. Fossils are not available, and even if they were it would be impossible to determine all of the subspecific development in *oregona* because color is rarely preserved in fossils. As a result indirect evidence must be used. This is provided by a consideration of the effects Pleistocene climatic changes may have had on bringing about subspeciation in *oregona*.

It seems that isolation of groups of populations of *oregona* occurred at different times in relation to climatic changes. In southern portions of the species range populations were separated from each other during interglacial periods because of the formation of deserts. Northern incipient subspecies were probably separated during glacial times.

Because *oregona* is riparian, regions void of river systems and lakes act as geographic barriers. The great deserts of southwestern United States prove to be barriers (fig. 18), and distribution of subspecies is closely linked to wet and cool areas. Consequently it may be deduced that isolation and subsequent genetic divergence of southern populations took place when southwestern United States was largely desert; perhaps during the last interglacial period. During glacial times, on the other hand, river systems were very extensive and many lakes occurred in the southwest (Blackwelder 1948, Hubbs and Miller 1948). In these regions populations were undoubtedly dispersed most widely in glacial times and presumably gene flow was uninhibited.

Conversely, partitioning of incipient subspecies that existed in northern regions of this species range probably occurred during glacial times, while range expansions occurred in interglacial periods. In glacial periods great ice masses moved down from the north, scarcely crossing the Canadian-American border in the west. These undoubtedly obstructed gene flow between aggregates of populations on the eastern portions of the Rockies and populations further west by way of northern United States and southern Canada. For example if an ice mass was at

present established across the northwest *o. oregona* and *o. guttifera* would be spatially isolated because they normally intergrade in Idaho, Montana, southern Canada and northern Utah. Similarly glaciers that developed throughout most of the major mountain ranges in glacial times must have reduced east-west gene flow further south.

Interpretation of present distribution of the subspecies and knowledge of the events of the Pleistocene epoch suggests the following course of subspeciation (see fig. 32). Two subspecies of *C. oregona*, *oregona* and *guttifera* as they are defined here, were formed in part during the Iowan glacial stage. A uniform "protooregona" species was distributed across northwestern United States and southwestern Canada prior to this period. With the advent of the Iowan ice mass and glacier formation all "protooregona" populations north of the Canadian - American border were probably annihilated, at least as a result of cooling and, two large populations were isolated, one on either side of the Continental Divide; race A on the west and race B on the east. Both races were more widespread in the south than they now are. Race B occupied all of the Great Basin, Arizona and regions west of the Sierra Nevada. Geographic variation was pronounced in this race with brown forms predominant in the north and blue in the south. Restricted to regions east of the Continental Divide in the north, race A extended southward into Colorado and New Mexico then swung northward through lower elevations in northwestern New Mexico and northeastern Arizona and eastern Utah. This was the situation when the Prairie interglacial stage began.

Much division and spatial isolation between southern populations took place during the Prairie interglacial as a result of vast desert formation. During this stage southern, blue populations of race B subspeciated to *maricopa* and brown western (race B) populations to *oregona*. Race A forms became *navajoensis* and *guttifera*. Distributions of aggregates of populations shrank and assumed geographic areas approximately where the subspecies of *C. oregona* now exist. *C. oregona* blue populations in the south were pinched off from their counterparts in the northwest by deserts where the Mohave Desert and Great Basin are now located. In Utah *navajoensis* populations remained partially isolated from populations in Colorado by the intervening Rocky Mountains, and were isolated from the effects of the *oregona* blue forms in the southwest. In the north *oregona* and *guttifera* reinvaded regions south of the retreating ice mass in northern United States and Canada and formed marked hybrid areas wherever their ranges came into contact.

Southern hybrid zones were primarily formed in the Wisconsin glacial period as a result of expanding subspecific ranges. Pluvial lakes that were reestablished in desert areas along with revived river systems served as routes for expanding ranges and intergradation was widespread. In the north *oregona* and *guttifera* were isolated from each other as in the previous glacial period.

Since the Wisconsin ice age ranges of subspecies have shrunk in the southwest. A few specimens phenotypically *maricopa* have been found in southern California but true *maricopa* is abundant only in central Arizona. This implies that *maricopa* once was more extensively distributed. On the other hand *oregona* and *guttifera* ranges appear to be expanding in the north.

Furthermore distribution of hybrids in the southwest has recently been
reduced in area. Evidence of this is available in southeastern Arizona
and southwestern New Mexico. Pure *maricopa* specimens along with
maricopa x *navajoensis* hybrids exist in these regions, but pure populations
of *navajoensis* are located in northeastern Arizona and northwestern New
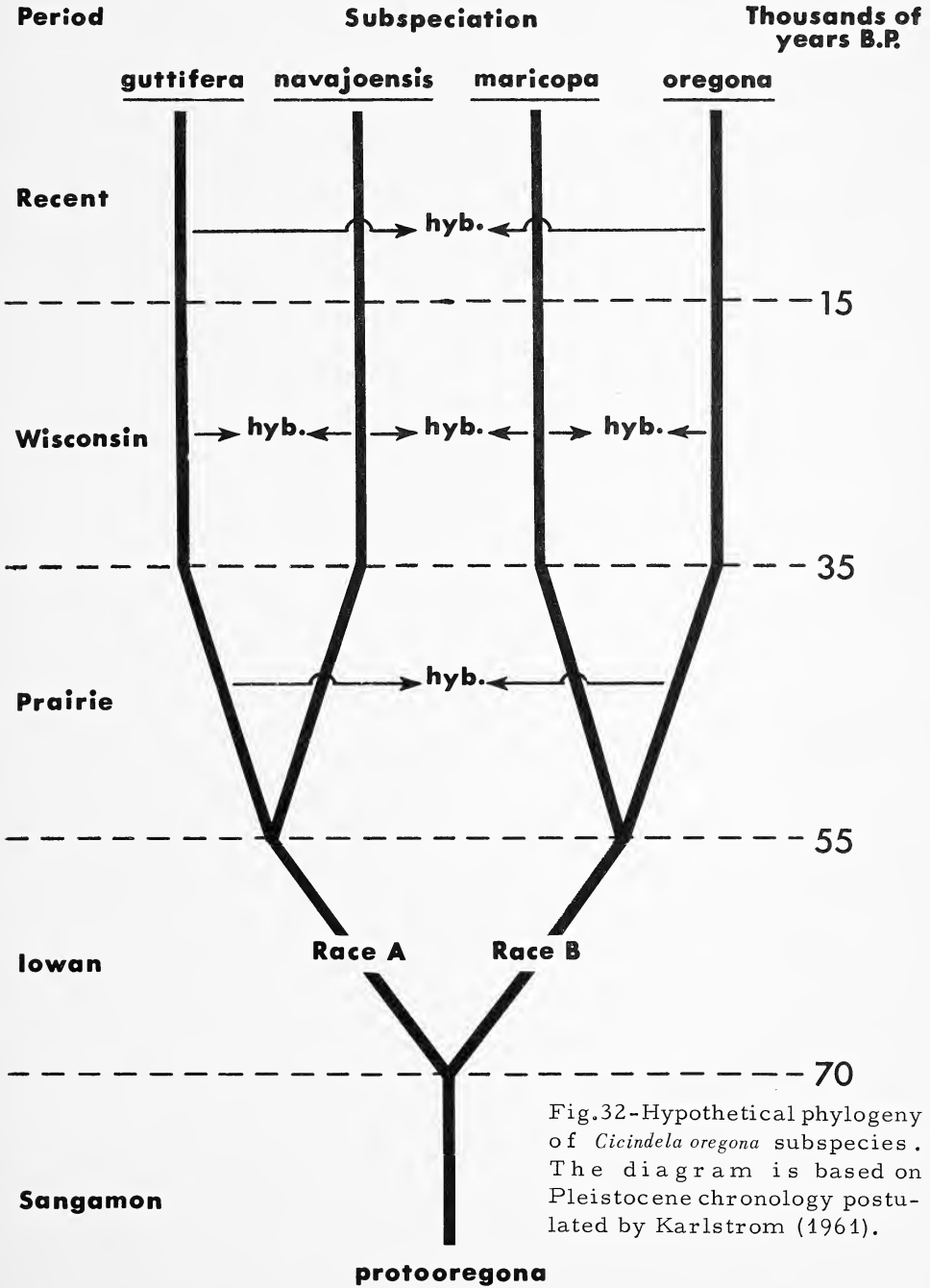Mexico, many miles away from where the hybrids are found.

Fig.32-Hypothetical phylogeny
of *Cicindela oregona* subspecies.
The diagram is based on
Pleistocene chronology postu-
lated by Karlstrom (1961).

Cicindela duodecimguttata and *C. o. guttifera* may have formed hybrid populations along the southeastern foothills of the Rocky Mountains during Pleistocene times. Such hybrid populations were probably subject to extreme fluctuations as in Nordegg, Alberta at the present time (p. 156). These unstable intermediate forms had no profound effect on the parental forms.

This discussion presents one interpretation of subspeciation in *C. oregona* that is based on knowledge of distribution pattern of *C. oregona* and Pleistocene events in southwestern United States. Undoubtedly other explanations of the available data are possible.

Distribution

I examined 6,073 specimens. Several specimens appeared to be labelled wrongly. Two *maricopa* specimens were labelled Fort Garland, Castilla County, Colorado. Fort Garland is well into *guttifera* territory beyond the northern limits of *maricopa* at that longitude. Another *maricopa* specimen is labelled Sonoma County, California, which is in northwestern California approximately seven hundred miles north of the *maricopa* specimens in San Diego. Five specimens of *o. guttifera* were labelled as being collected in Santa Rita Mountains. This is unlikely but not impossible since these specimens could represent a relict population which has survived in these mountains since the end of the Pleistocene.

Cicindela oregona oregona LeConte. Canada. BRITISH COLUMBIA:
Abbotsford, 9; Agassiz, 25; Ainsworth, 6; Aiyansh, 1; Atbara, 7; Bear Foot, 1; Cherrierville, 2; Chilliwack, 2; Columbia Lake, 1; Comox, 8; Copper Mine, 2; Courtenay, 4; Cranbrook, 3; Creston, 139; Duncan, 3; Duncan and Cowichan Lake, 1; Elk Lake, 1; Fairmont, 1; Field, 1; Fraser River, 1; Gabriola, 2; Garibaldi Park, 29; Golden, 1; Goldstream, 1; Harrison, 2; Hatzic, 20; Hope, 1; Howser, 7; Huntingdon, 15; Kamloops, 3; Kaslo, 8; Keremeas, 2; Likely, 1; Lillooet, 9; Lynn Valley, 1; Lytton, 2; Mabel Lake, 2; MacGillivray, 53; McIntyre Creek, 1; Merritt, 22; Miracle Beach, 60; Misson City, 20; Nanaimo, 1; Nanoose, 5; North Bend, 4; Okanagan Lake, 1; Oliver, 61; Osoyoos, 2; Peachland, 3; Pender Harbour, 5; Penticton, 3; Point Grey, 2; Powell River, 2; Radium, 3; Read Bay, 4; Riondel, 11; Rock Creek, 7; Salmon Arm, 3; Sanca, 12; Shuswap Falls, 1; Stillwater, 1; Summerland, 4; Toad Inlet, 1; Trinity, 1; Vancouver, 79; Vancouver Island, 5; Vaseaux Lake, 13; Vernon, 42; Victoria, 18; White Lake, 2; Windemere, 1; Wynndel, 8.
    United States. CALIFORNIA: Alameda County: 11; Berkley, 17; Oakland, 10. Alpine County: 7; Soda Springs, 1. Butte County: Chico, 4; Chico Creek, 1; Oroville, 2. Calaveras County: Big Trees, 6; Mokelumne Hill, 7. Contra Costa County: Antioch, 32. Del Norte County: Crescent City, 6; Terwah, 1. Eldorado County: Bijou, 2; Desolation Valley, 1; Echo, 1; Fallen Leaf Lake, 1. Fresno County: Cedar Grove, 1; Coalinga, 12; Fresno, 3; Huntington Lake, 15; Macy Mines, 1; Stevenson Creek, 1. Glenn County: Elk Creek, 2. Humbolt County: Blue Lake, 2; Blairs Ranch, 3; Bridgeville, 28; Bridgeville (15 miles east), 5; Eel River, 1; Eureka, 9; Ferndale, 2; Fort Seward, 26; Hoopa, 44; Miranda, 29; Pepperwood, 1; Jct. Redwood Creek and Rte. 299, 12; Scotia, 3; Trinidad, 1; Trinidad (5 miles south), 25; Van Duzen River, 2; Willow Creek, 1. Inyo County: Bigpine (2 miles west), 12; Bigpine (2 miles north), 20; Biship, 2; Lone Pine, 11; Olancha, 18; Owens Lake, 1. Kern County: Bakersfield, 2; Bakersfield (4 miles east), 9; Cottonwood Creek, 1; Isabella, 1. Lake County: Borax Lake, 5; Hullville, 3; Lake Pillsbury, 8; Lakeport, 2; Lower Lake, 1. Lassen County: Facht, 1; Gournaz, 4; Madeline, 1. Pine Creek, 1; Susanville, 1. Los Angeles County: Burbank, 2; Covina, 1; Crystal Lake, 1; El Monte, 3; Gabriel Mountains, 1; Los Angeles, 57; Palmdale, 1; Point San Pedro, 5; San Pedro, 1; Santa Monica, 10; Tropico, 1. Madera County: Jackass Meadow, 1; Madera, 1; North Fork, 2; Placer Station, 2. Marin County: Bon Tempe, 4; Dillon Beach, 6; Inverness, 1; Lagunitas, 1; Mill Valley, 1; Point Reyes Station, 4. Mariposa County: Mariposa Grove, 1; Yosemite National Park, 22; Yosemite Valley, 1. Mendocino County: Caspar, 1; Eagles Nest, 1; Fort Bragg, 1; Littleriver, 2; Philo, 1; Yorkville, 3. Merced County: Lake Merced, 7; Los Bonos, 1; Santa Rita, 2. Modoc County: Cedarville, 2; Goose Lake, 1; Lake City, 1. Mono County: Coleville, 1; Lake Mary, 5; Mono County, 13; Mono Lake, 6; Sonora Pass, 2; Topaz Lake, 8. Monterey County: Bradley, 1; Carmel, 34; Pacific Grove, 1; Salinas River, 6; Soledad, 2; Stone Canyon, 4. Nevada County: Norden, 2; Soda Springs, 1; Truckee, 7. Orange County: Anaheim, 5; Laguna Beach, 2. Placer County: Brockway, 2; Dutch Flat, 2; Lake Tahoe, 75; Summit, 1. Plumas County: Bucks Ranch, 20; Clio, 1; Meadow Valley, 1; Near Twain, 1; Quincy (4 miles west), 2; Tobin, 1; U.S. Alt. 40 (2.1 miles W., U.S. 395), 30; Walker Mine, 1. Riverside County: Hemet Res., 8; Idyllwild, 20; Indio (30 miles north), 2; Lake Elsinore, 2; Riverside, 12; San Jacinto Mountains, 13; Temecula, 1; White Water Canyon, 2. Sacramento County: Sacramento, 19. San Bernadino County: Big Bear, 7; Big Bear Valley, 1; Cajon, 3; Cajon Pass, 1; Colton, 21; Victorville, 13; Yermo, 1. San Diego County: Descanso, 1; Guatay, 1; Julian, 3; Mesa Grande, 5; Oceanside, 1; Poway, 1; Warner Springs, 1. San Francisco County: San Francisco, 22. San Joaquin County: Stockton, 7. San Luis Obispo County: Paso Robles, 1; Pismo Beach, 3. San Mateo County: Salada Beach, 7. Santa Clara County: Los Gatos, 1; Palo Alto, 2; San Jose, 3. Santa Cruz County: Santa Cruz, 4; Sequel Basin, 3. Santa Cruz Island, 13. Shasta County: Bumpas Hell, 1; Lassen National Park, 5; Castella, 2; Hat Creek, 3; Hat Lake, 1; Manzanita Lake, 4; Mount Lassen, 3; Redding, 3; Shasta National Forest, 3; Shasta Springs, 11; Viola, 1. Sierra County: Sierravilla, 9; Yuba Pass, 1, Siskiyou County: Dillon Creek, Klamath River Valley, 21; Dunsmuir, 4; Gottville, Klamath River Valley, 24; Etna, 3; McCloud, 10; Tule Indian Reservation, 1. Solano County: Dixon, 1. Sonoma County: Cloverdale, 15; Duncans Mills, 1; Santa Rosa, 4. Stanislaus County: Patterson, 1. Sutter County: Yuba City, 5. Tehama County: San Bernardino Mountains, 1. Trinity County: Douglas City, 14; Jct. Mad River, Rt. 36, 37. Tulare County: Burton Meadow, 1; Kaweah, 2; Lemoncove, 6; Lloyd Meadow, 1; Sequoia National Park, 8. Tuolumne County: Hardin Flat, 12; Pinecrest, 1; Strawberry Resort, 11. Yolo County: Davis, 6. Localities of unknown counties: Alta, 1; Asilomar, 1; Bear Lake, 1; Bentons Crossing, 3; Big Dalton Dam, 6; Borego, 1; Bryson 1; Carrville, 14; Charity Valley, 1; China Flat, 2; Colony Mill, 1; Ebbec Pass, 1; El Mirador, 1; Fort Ross, 3; Giant Forest, 2; Hackamore, 5; Hartsooks, 2; Hickey Grove, 7; Hueneme, 1; Ingleside, 2; Kings River, 11; Lake Clay, 1; Lassen Creek, 1; Leech Lake Mountain, 6; Marble Bridge, 1; Marwedel, 1; Miami, 1; Mount Tallac, 1; Panoche Canyon, 1; Paradise Springs, 1; Pohono, 3; Poloa, 4; Poso Creek, 1; Redstone, 2; Sandflat, 6; San Juan, 1; San Pedro Valley, 15; Shaver, 1; Sheep Creek, 1; Silliman, 1; Simmler, 1; Sylvania, 1; Tassajara, 5; Towie, 1; Tunnel R.S., 1; Vera Lake, 1; Waddell Beach, 2; Walker, 6. IDAHO: Ada County: 6; Boise, 2. Blaine County: Carey, 1. Boise County: 13. Bonner County: Hope, 1; Priest Lake, 1; Priest River, 5; Sandpoint, 1. Boundary County: Selway Falls (2.5 miles south), 1. Canyon County: 8; Nampa, 1. Elmore County: Atlanta, 2. Gooding County: Hagerman, 1. Idaho County: 4. Jerome County: Jerome, 1. Kootenai County: Cataldo, 1; Coeur d'Alene, 1. Latah County: Moscow, 7; Troy, 1. Nez Perce County: Lake Lowell, 2; Lewiston, 5. Owyhee County: Homedale, 1; Owyhee County, 62. Power County: American Falls, 1. Shoshone County: Wallace, 1. Valley County: Cascade, 1; McCall, 1. Valley County: 20. NEVADA:

Churchill County: Fallon, 3. Douglas County: Lake Tahoe, 1; Minden, 24. Elko County: Elko, 4; Lamoille, 1. Mineral County: Hawthorne, 10; Walker Lake, 45. Ormsby County: Carson City, 3. Pershing County: Lovelock, 1. Washoe County: Gerlach, 2; Mount Rose, 1; Nixon, 1; Pyramid Lake, 18; Reno, 19; Verdi, 1. White Pine County: McGill, 1. OREGON: Baker County: Pine Creek, 1; Richland, 1. Benton County: Corvallis, 8; Umatilla, 10. Clackamas County: Estacada, 4. Clatsop County: Cannon Beach, 1; Clatsop Beach, 2. Columbia County: Rainier, 1. Coos County: Cape Arago, 2; Charleston, 16; Coos Bay, 4; Coos Head, 1. Curry County: Humbug Mountain, 6; Port Orford, 12. Grant County: John Day George, 2. Harney County: Frenchglen, 8; Malheur Lake, 15; P. Ranch, 1; Steens Mountains, 15. Hood River County: Hood River, 8; Mount Hood, 2; Parkdale, 1. Jackson County: Medford, 10; Rogue River, 1; Ruch, 2. Josephine County: Grants Pass, 5; Hells Gate Bridge, 2; Murphy, 1. Klamath County:Crater Lake, 2; Klamath Lake, 6; Lake O Woods, 3; Pinehurst (21.9 miles east), 1. Lake County: Lake Albert, 12; Paisley, 1. Lane County: Eugene, 13; Florence (3 miles north), 13. Lincoln County: Depoe Bay, 2; Newport, 6; Waldport, 5; Yachats (5 miles south), 5. Linn County: Cascadia, 1. Malheur County: Sucker Creek Canyon, 1. Marion County: Detroit, 1. Multnomah County: Portland, 9. Tillamook County: Pacific City, 1; Tillamook, 1; Woods, 4. Umatilla County: Echo, 2; Hermiston, 3; Meadow Lake, 1. Wasco County: The Dalles, 7; Tygh Valley, 1. Yamhill County: Dayton, 6; McMinnville, 4. Localities of unknown counties: Alvord Hot Springs, 3; Blitzen Valley, 1; Boiler Bay, 5; Buell, 1; Devils Lake, 1; Durnep, 2; McNair Lake, 1; Moffat Mead, 1; Ocean Park, 1; Oregon (south east), 3; Santiam, 4; Sparks Lake, 1; Whitman, 2. UTAH: Salt Lake County: Alta, 13; Brighton, 1. Utah County: American Fork Canyon, 9. WASHINGTON: Adams County: Othello, 7. Asotin County: Asotin, 1; Clarkston, 1. Benton County: Paterson, 1. Chelan County: Leavenworth, 1; Peshastin, 4; Stehekin, 1; Wenatchee, 5. Clallam County: Port Angeles, 1. Columbia County: Huntsville, 3. Douglas County: Moses Coulee, 3. Franklin County: Kahlotus, 2; Pasco, 1. Grant County: Beverly, 3; Goose Lake,1; Moses Lake, 1; Stratford, 9. Grays Harbor County: Moclips, 3. Island County: Coupeville, 1; Whidby Island, 53. Jefferson County: Port Townsend, 17. King County: Auburn, 2; Bothell, 4; Cedar Mountain, 5; Maple Valley, 3; Renton, 13; Seattle, 80; Selleck, 1; Snoqualmie, 1. Kitsap County: Bremerton, 49; Chico, 34; Gorst, 120; Keyport, 1; Kingston, 13; Manchester, 1. Kittitas County: Ellensburg, 2; Vantage, 12. Klickitat County: Goldendale, 1; Goldendale (32.3 miles north), 22. Lewis County: Chehalis, 2. Lincoln County: Sprague, 2. Mason County: Lake Cushman, 1; Spillman, 2. Okanogan County: Brewater, 5. Pacific County: Bay Center, 4; Ilwaco, 1; Long Beach, 1; Nahcotta, 2; North Cove, 1; Ocean Park, 3. Pend Oreille County: Newport, 1. Pierce County: Buckley, 1; Chinook Pass, 3; Mount Rainier, 1; Summer,1; Tacoma, 4. San Juan County: False Bay, 1; Friday Harbor, 9. Skagit County: Anacortes, 18. Snohomish County: Cicero, 2; Darrington, 6; Everett, 3; Index, 4; Sulton, 1; Verlot, 8. Spokane County: Medical Lake, 4; Spokane, 13. Stevens County: Wellpinit, 1. Thurston County: Olympia, 1; Tenino, 5. Walla Walla County: Dixie, 10; Lowden, 1; Touchet, 27; Wallula Gap, 1. Whatcom County: Bellingham, 1; Mount Baker, 2. Whitman County: Almota, 3; Pullman, 30; Wawawai, 11. Yakima County: Toppenish, 4; White Swan, 1; Yakima, 2. Localities of unknown counties: Barkerville, 1; Blue Mountains, 1; Central Ferry, 1; Clifton, 1; Ginko State Park, 2; Half Moon Lake, 2; Lyone Ferry, 9; Neppel, 10; Paha, 5; Pot Holes, 2; Saratoga Beach, 5; Silverton, 1; Skating Lake, 1; Stillaguamish, 2; Tolsak, 1.

Cicindela oregona guttifera LeConte. Canada. ALBERTA:
Kootenay Plains, 14. BRITISH COLUMBIA: Aiyansh, 1; Blue River, 1; Bucks Bar, 1; Cariboo Road (mile 185), 2; Glenora, 1; McNab Creek, 1; Juskalta, 6; Massett, Queen Charlotte Islands, 10; Moresby Camp, Queen Charlotte Islands, 9; Queen Charlotte City, 1; Stickeen River, 1; Tlell, Queen Charlotte Islands, 9. NORTHWEST TERRITORIES: Fort Good Hope, 2; South Nahanni River, 1. YUKON TERRITORY: Kirkman Creek, 1; Watson Lake, 6.

United States. ALASKA: Eagle, 3; Fairbanks, 5; Fort Yukon, 11; Haines, 12; Tanana River, 27; Valdez, 4; Yukon River, 1. COLORADO: Alamosa County: Alamosa, 3. Boulder County: Boulder, 5; Jamestown, 2; Lyons, 1; Pinecliffe, 2. Chaffee County: Buena Vista, 2; Salida, 3. Clear Creek County: Georgetown, 1. Conejos County: Cumbres Pass, 4; La Manga Pass, 4. Douglas County: Larkspur, 2. El Paso County: Cascade, 1; Colorado Springs, 6; Colorado Springs (10 miles south), 6; El Paso County: 2; Manitou Springs, 5. Freemont County: Coalcreek, 1. Garfield County: Glenwood Springs, 1. Grand County: Big Muddy Creek, 1; Fraser, 1. Jefferson County: Golden, 7. La Plata County: Electra Lake, 1. Larimer County: Estes Park, 25; Fort Collins, 6. Las Animas County: Trinidad, 26. Mineral County: Creede, 4; Wolf Fall Creeks, 2. Ouray County: Ouray, 1. Park County: Colorado Springs (50 miles west), 4. Pitkin County: Aspen, 4. Saquache County: Great Sand Dunes National Monument, 4. Teller County: Victor, 1. Localities of unknown counties: Berkley, 1; Gothic, 1; Rockwood, 2; South Fork, 1; Thomasville, 1. NEW MEXICO: Bernalillo County: Albuquerque, 1. San' Doval County: Bernalillo, 1; Jemez Mountains, 17; Jemez Springs, 47; Jemez Springs (9 miles north), 73; Jemez Springs (10 miles north), 43. San Miguel County: Beulah, 2. Santa Fe County: Pecos River, 17; Santa Fe (3 miles east), 2. Localities of unknown counties: San Antone, 1. UTAH: Beaver County: Beaver Creek, 1. Cache County: Logan, 3. Juab County: Levan (5 miles south), 1. Millard County: Lynndyl, 1. Piute County: Piute Reservoir, 18. Rich County: Bear Lake, 1. Salt Lake County: Mount Dell Creek, 1; Parley Canyon, 3; Salt Lake City, 18; Salt Lake County, 24. Sampete County: Sevier Bridge Reservoir, 21. Summit County: Echo, 2; Park City, 3. Tooele County: Stockton, 15. Uintah County: Power Plant, 2. Utah County: Mount Timpanogos, 20; Payson, 1; Provo, 17; Provo Canyon, 26; Utah Lake, 19. Wasatch County: Soldier Summit, 3; Wasatch County, 2. Weber County: Ogden, 13; Ogden (30 miles east), 8. Localities of unknown counties: Emigration, 1; Hillneck Canyon, 1; Kawara, 6; Red Butte Canyon, 1; Salt Creek Canyon, 1; Silver Lake, 5; Vineyard, 26. WYOMING: Albany County: Centennial, 1; Jelm, 88. Carbon County: Baggs, 2; Saratoga (8 miles south), 24. Fremont County: Lander, 1. Lincoln County: Labarge (11 miles south), 21. Sublette County: Big Sandy Reservoir, 11; Half Moon Lake, 11; Sweetwater River, 2. Sweetwater County: Green River, 54; Green River (26 miles south), 6; Old Ford on Green River, 34; Sweetwater County, 7. Uinta County: Fort Bridger, 64; Lyman, 4.

Cicindela oregona maricopa Leng. United States. ARIZONA:
Coconino County: Grand Canyon, 1. Gila County: Globe, 56; Roosevelt Lake, 4; San Carlos, 1; Sierra Ancha Mountains, 7. Graham County: Rylas, 8. Greenlee County: Clifton, 9. Maricopa County: Phoenix 25 ; Tempe, 1. Pima County: Tuscon, 5. Pinal County: Pinal Mountains, 7. Yavapai County: Cottonwood, 1; Haslampa, 3; Prescott, 266. Localities of unknown counties: Bad Creek Canyon, 1; Bradshaw Mountains, 1; Mogollon Mountain, 1; Oak Creek Canyon, 2. CALIFORNIA: Los Angeles County, 1. San Bernardino County: Barstow, 1. NEVADA: Lincoln County: Caliente, 13; Meadow Valley, 1. NEW MEXICO: Otero County: Cloudcroft, 2; Mountain Park, 15. UTAH: Iron County: Cedar City Canyon, 2.

Cicindela oregona navajoensis Van Dyke. United States.
ARIZONA: Navajo County: Betatakin, 9; Kayenta, 25; Navajo Mountain, 6. Moffat County: Echo Park, 2; Massadona, 4. Montezuma County: Four Corners, 4. UTAH: Grand County: Floy, 40; San Juan County: Blanding (10 miles west), 4; National Monument, 1; Navajo Mountain Trading Post, 7.

Canada. ALBERTA: Banff, 52; Laggan(Lake Louise), 22; Waterton Park, 5. BRITISH COLUMBIA: Athalmer
24; Canal Flats, 8; Cinema, 7; Fernie, 25; Fort Fraser, 13; Kootenay National Park, 6; Moyie, 2; Terrace
35; Wasa, 7; Yoho National Park, 17.
        United States. IDAHO: Bannock County: Pocatello, 7. Bear Lake County: Bear Lake, 119
Bloomington Lake, 10. Franklin County: Franklin Basin, 11. Fremont County: Parker, 2. Lemhi County
Salmon (21 miles north), 6. MONTANA: Cascade County: Great Falls, 2. Flathead County: Hungary Horse
11; Kalispell, 1. Gallatin County: Bozeman, 2; Gallatin County, 2; Gallatin River and Highway 10, 14;   Lak
Hebgen, 12; Missouri River (headwaters), 2; Three Forks (3 miles west), 13. Glacier County: Lower Medicin
Lake, 66. Lewis and Clark County: Craig, 14; Helena, 50; Hardy (15 miles south west), 33. Lincoln County
Troy, 1. Missoula County: Frenchtown, 3. Park County: Gardiner (5 miles north), 86. Ravalli County:
Darby, 2; Florence, 2; Hamilton, 3; Ravalli County, 2; River Bottoms, 10; Skalkaho, 1. Sanders County:
Whitepine, 1. Silver Bow County: Butte, 1. Localities of unknown counties: Beaver Creek, 8; Bitter Root
Mountains, 2; Lost Horse Canyon, 1; Marias River, 2; Stickney Creek, 34. WYOMING: Teton County:
Black Rock Creek, 4; Grand Teton National Park, 13; Hobach Canyon, 3; Jackson Hole National Monument, 27;
Moran (38 miles east), 46. Yellowstone National Park, 140.

United States. CALIFORNIA: San Diego County: San Diego, 45.

United States. UTAH: Beaver County: Beaver (4 miles east), 11; Beaver Canyon, 6; North Creek, 1. Iro
County: Burkshire, 1; Cedar City, 9; Iron Springs, 2; Parowan (5 miles south east), 7; Parowan Canyon, 12.
Kane County: Glendale, 1; Kanab, 1; Orderville, 2. Washington County: Bellvue, 2; Pine Valley, 2; Pintura
3; Saint George, 8; Santa Clara, 3; Zion National Park, 26. Localities of unknown counties: Mount Meadows
1; Weeping Rock, 1.

United States. COLORADO: Mesa County: De Beque, 4; Palisade, 7. NEW MEXICO: McKinley County
Fort Wingate, 33. San Juan County: Farmington, 3. UTAH: Uintah County: Dinosaur National Monument
1; Vernal, 12.

United States. ARIZONA: Apache County: White Mountains, 8. Cochise County: Chiricahua Mountains, 2.
Navajo County: Carrizo, 8; Cibeque Creek, 2. NEW MEXICO: Catron County: Luna, 5. Brant County
Silver City, 6.

The Species *Cicindela depressula* **Casey.**

Cicindela depressula depressula Casey 1897:297. Type locality - Placer County,
    California. Leng 1902:150. Rivalier 1954:253.
        Cicindela oregona depressula Hatch (not Casey 1897) 1953:42. Horn
        1930:82. Wallis 1961:24.
Cicindela depressula eureka Fall 1901. Type locality - Humbolt County, Calif-
    ornia. NEW COMBINATION.
        Cicindela eureka Fall 1901:307.   Leng 1902:149.   Horn 1930:82.
        Rivalier 1954:253.

        Two constant differences set apart *depressula* from other species
of the *maritima* group. First, in *depressula* two or three hairs usually occupy
the small area near the front inner edge of each eye; four hairs are seldom
present. If these hairs are abraded, setigerous punctures indicate
their former positions. Second, the distal end of the median lobe of the
male genitalia of *depressula* has two distinct, broad flanges that form a
blunt apex (for details see fig. 3, and p. 91). A partially diagnostic
character is the form of the middle band of the elytra. In Oregon and
California the middle band of *depressula* tapers evenly posteriorly. This
contrasts with the sharp bend in the middle band of *oregona*. On the other
hand in Washington, Canada, and Alaska the middle band of *depressula*
often appears identical with that of *oregona*.
        I collected *oregona* and *depressula eureka* on the same sand bank along
the Van Duzen River near Bridgeville, California; and Ball (personal
communication) collected *d. depressula* and *o. guttifera* in the same area at
Terrace, British Columbia, and at Haines, Alaska. There was no evidence
of hybridization or cross-mating at these locations, and this suggests
that *depressula* and *oregona* are specifically distinct in spite of their many
shared characteristics.

Notes on Synonymy

The tiger beetles called *depressula* and *eureka* are very similar to one another, differing mainly in color and markings of the elytra and in seasonal occurrence of adults. They are also allopatric. The differences, however, are not absolute; that is, range of variation in the diagnostic characters of the two forms is slightly overlapping. Furthermore, Rumpp has from the Olympic National Forest, Washington, a series of specimens interpreted as hybrids between *depressula* and *eureka*. These considerations of variation and distribution suggest that *depressula* and *eureka* are conspecific, but are subspecifically distinct. Rumpp and I have reached this conclusion independently.

Geographic Variation and Subspecies

Eleven population samples, whose geographic positions collectively span the known range of this species were selected on the basis of geographical location and number of specimens and were examined for variation. Elytral size, elytral color, pleural color, and the condition of the middle band vary. There may be a humeral dot on the elytra. Variation in these was analyzed and the results are summarized in tables 6-8.

Length of elytra was examined in a cursory fashion (table 6). Mean values for females are higher than those for males from the same locality. Mean values for samples from lowland, coastal regions in Humbolt County, California are normally higher than mean values for populations from Mount Rainier, Washington. This is the reverse of the pattern of the size variation in *oregona*.

TABLE 6 - Variation in length of elytra of male and female *d. depressula* and *d. eureka*.

	Sex	N	Range mm	$\overline{X}$ ± SE	SD	CU
d. depressula	male	25	6.62-8.07	7.25 0.07	0.35	4.85
	female	25	6.70-8.42	7.60 0.08	0.38	5.00
d. eureka	male	25	6.72-8.12	7.51 0.06	0.32	4.21
	female	25	7.35-9.03	8.31 0.08	0.42	5.02

Populations of the Cascade Range differ in phenology from those of the Pacific coast of Oregon and California. At these latitudes populations from these regions do not meet, for apparently there is a difference in seasonal occurrence of adults. Most adults in alpine regions are active during the middle and late summer but in coastal populations the adults are out earlier in the year. On a collecting trip in June 1963 *depressula* was not found at high elevations, but specimens of

coastal populations were collected in northwestern California.

Elytra of *depressula* have a metallic lustre and are either brown, green or blue (table 7). There is no evidence of a uniform character gradient. Brown elytra are prevalent in coastal regions of northern California, on Mount Hood, Oregon and on Mount Rainier, Washington. Specimens with green elytra occur throughout the range of the species, but are most frequent at both northern and southern extremities of the range, e. g. Haines, Alaska and Eldorado County, California. Blue elytra are most common in populations from Washington and southern British Columbia and as such parallel those of *oregona* in southwestern British Columbia. In California, over 90 per cent of specimens from Cascade Range localities are green whereas over 90 per cent from the coast are brown.

Color of thoracic pleura is rather variable in every population listed in table 7 except for Lassen National Park and Eldorado County, California. Specimens with green and coppery thoracic pleura are more common than those with blue.

Data on variation in the humeral dot are presented in table 8. The humeral dot is present in all specimens collected in lowland localities near the Pacific coast. Among specimens from localities in the Cascade Mountains, the dot may be present or absent. Over 90 per cent of the specimens from north of and including Mount Baker, Washington have humeral dots on the elytra, but only 10 per cent of the specimens from southern Oregon and California have them. In the Mount Rainier, Washington and Mount Hood, Oregon samples the two conditions occur in about equal frequencies. The variation appears to be clinal.

The middle band of the elytra may be broken or complete, and the data on the frequencies of these conditions in various population samples are presented in table 8. In general the broken condition of the middle band is more common southward, among montane populations, but this is a poorly marked trend. All of the specimens from coastal regions in California have a complete middle band.

Variation in hair between the eyes is not tabulated. However, between the eyes, in the middle of the head, one to four very fine hairs are present in coastal specimens' from southern localities. The hairs rarely appear on specimens from northern montane regions, or on specimens from montane localities in California. These hairs should not be mistaken for the setae near the front inner edge of the eyes that are characteristic of *depressula* as a whole.

Most northern populations studied exhibit appreciable variation. The five specimens from Haines, Alaska are less variable but more material is necessary from northern localities to obtain a better knowledge of the degree of variation. In the central portions of the range, variation is discordant. Southern populations, however, are remarkably uniform.

Of the characters considered above, specimens collected in southern areas of the Cascade Range vary mainly in the condition of the middle band. Almost all of these specimens have the elytra and thoracic pleura green, and no humeral dot. These characters are generally most common in northern populations.

Mountain populations of California and Oregon are distributed in a thin band along the Cascade Range where they are confined to high altitudes, but further north they also occur in the Rockies (Mount Revelstoke, British Columbia) and altitudinal preference is not as marked as in southern regions (fig. 33).

TABLE 7 - Color variation of elytra and thoracic pleura of population samples of *Cicindela depressula*.

	Color of Elytra			Color of Pleural Sclerites		
	Brown	Green	Blue	Copper	Green	Blue
	N	N	N	N	N	N
d. depressula						
Haines, Alaska	1	4	0	4	1	0
Garibaldi Park, B. C.	1	7	7	8	6	1
Mount Baker, Wash.	1	5	6	2	6	4
Mount Rainier, Wash.	201	115	135	200	208	43
Mount Hood, Oregon	40	5	0	33	12	0
Crater Lake, Oregon	8	45	6	9	44	6
Lassen Nat. Pk. , Calif.	0	22	1	1	21	1
Eldorado County, Calif.	0	25	1	1	24	1
d. eureka						
Orick, Red Ck. , Calif.	12	0	0	3	5	4
Arcata, Mad River, Cal.	38	0	0	3	5	4
Van Duzen River, Calif.	41	5	0	5	31	10

TABLE 8 - The occurrence of elytral humeral dot and variation in the condition of the middle band of elytra among population samples of *Cicindela depressula*

	Specimens with dot/N	Specimens with complete band/N
d. depressula		
Haines, Alaska	5/5	4/5
Garibaldi Park, B. C.	14/15	13/15
Mount Baker, Wash.	11/12	12/12
Mount Rainier, Wash.	232/451	429/451
Mount Hood, Oregon	26/45	29/45
Crater Lake, Oregon	1/59	9/59
Lassen Park, Calif.	0/23	13/23
Eldorado County, Calif.	0/26	19/26
d. eureka		
Orick, Red. Creek, Calif.	12/12	12/12
Arcata, Mad River, Calif.	38/38	38/38
Van Duzen River, Calif.	46/46	46/46

Fig. 33.
Geographical
distribution of

C. d. depressula ●

C. d. eureka ◉
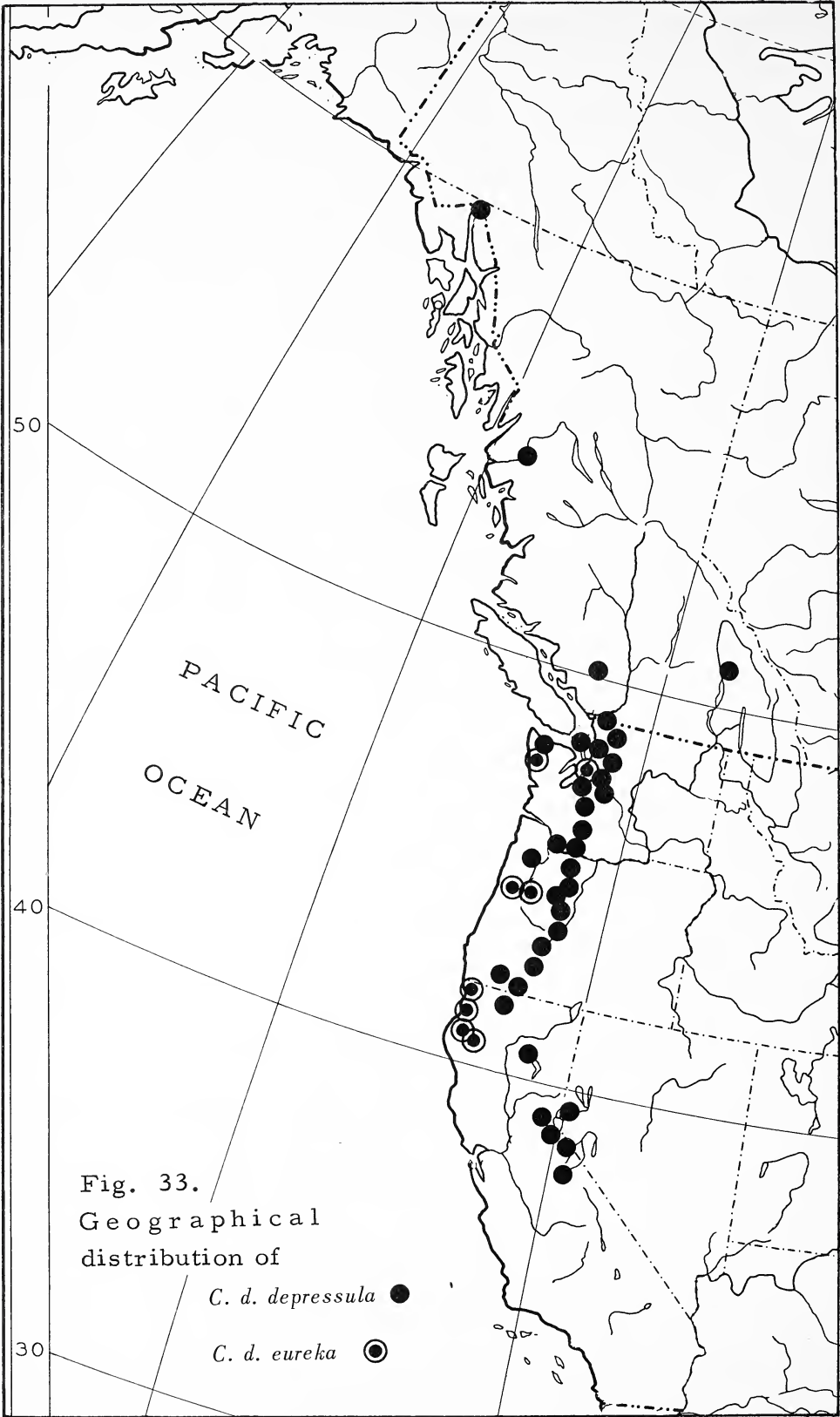
Coastal specimens from California differ in external characteristics from alpine individuals at the same latitude. Specimens from these lowland regions usually have brown elytra, a very long, usually complete middle band, a humeral dot, and coppery, green, or blue thoracic pleura. They occupy river banks near the Pacific coast of northern California, Oregon and Washington. Few specimens of this type have been collected in Oregon and Washington. They may not be abundant in these regions; or perhaps the adults come out only for a short period. On the other hand, more intense collecting in May and June may yield larger numbers of these forms from these areas.

These differences provide the basis for distinguishing two subspecies: a southern coastal one, *d. eureka* Fall and a mountain northern one, *d. depressula* Casey.

Variation in some morphological characters, phenology, hybridization, and distribution of *depressula* and *eureka* have been discussed above.

However, the apparently restricted distribution of *eureka* should be discussed further.

Distribution limits of *eureka* north of California are poorly understood because material is very scanty. In June 1963, a collecting expedition was made to the American southwest in order to obtain specimens of *depressula* and *oregona*. While in Humbolt County, California we obtained several large series of *eureka* near the mouths of Redwood Creek, and Mad and Van Duzen Rivers. Travelling from west to east in the Van Duzen River valley, we collected *eureka* as far east as Bridgeville which is approximately 1,000 feet above sea level and 30 miles east of the Pacific coast. Fifteen miles east of Bridgeville at an elevation of about 2,000 feet *eureka* was not present, nor was *eureka* present along Redwood Creek, 17 miles east of the Pacific coast. Thus the eastern limit of *eureka* is a short distance from the coast. At this latitude average temperature differences due to altitude may play a part in limiting *eureka* so closely to the coast. However, much ecological information is essential in order to understand the forces which confine *eureka* to such a restricted region in California.

C. depressula evolved in western North America probably as a cool-adapted lowland species, having shared a common ancestry with *duodecimguttata* . During a fairly recent glacial stage the range of the species was bisected by mountain glaciers with survivors to the west and to the east of the Cascades at low elevations. During this period of isolation differentiation occurred, with the coastal populations evolving the least in color pattern, but becoming bound to climatic conditions existing at lower elevations. This group became the subspecies *eureka* . As the glaciers retreated and the inland refugium became warmer and drier the populations isolated there (*d. depressula*) moved up the mountains or northwards or both ways. Differentiation then occurred in *d. depressula* with a loss of white markings in the southern members. In northern Washington the ranges of the two isolates met and hybridization took place.

Distribution patterns like that of *depressula* are evident in vertebrates such as *Sorex vagrans* Baird (Findley 1955), *Rana aurora* , Baird and Girard, and *Contia tenuis* Baird and Girard (Stebbins 1954).

Distribution

Of the 922 specimens of *depressula* examined, one appears to be incorrectly labelled Berkeley, Alameda County, California. This specimen does not resemble the subspecies *d. eureka* but it is phenotypically *d. depressula* the distribution of which is restricted to the Cascade Range at that latitude.

Cicindela depressula depressula Casey.   Canada.   BRITISH COLUMBIA: Diamond Head Trail, Garibaldi Park, 14; Jade Lake Trail, Mount Revelstoke, 1; Terrace, 4.
     United States. ALASKA: Haines Highway, near Haines, 5. CALIFORNIA: Alpine County: 7. Eldorado County: Echo Lake 15; Fallen Leaf Lake, 2; Keith Dome, 11; Mount Tallac, 10; Summit, 3. Mariposa County: Kerrick Meadows, Yosemite National Park, 2. Nevada County: Rucker Lake, 1. Placer County: Summit, 2. Shasta County: Kings Creek, Lassen Park, 6; Mount Lassen, 18. Siskiyou County: Walker, 3. Localities of unknown counties: Angora Park, 3; Carson Pass, 2; Charity Valley, 1; Sovoft, Sierra Nevada Mountains, 1; Warner Valley, 2. NEVADA: Washoe County: Mount Rose, 2. OREGON: Deschutes County: Three Creeks, 1; Todd Lake Meadows, 1. Douglas County: Diamond Lake, 1; Three Lakes, 1. Hood River County: Mount Hood, 54. Jackson County: Mount Ashland, 2. Jefferson County: Mount Jefferson, 1. Josephine County: Rogue Riffles, 1. Klamath County: Crater Lake, 60; Summit Lake, 2. Lake County: Linton Meadows, near Three Sisters area, 17. Lane County: Obsidian Trail, 1; Scott Lake, 9; Wikiup Plains, 5. Linn County: Big Lake, 1; Hoodoo, 2; Santiam, 1. Yamhill County: McMannville, 1. WASHINGTON: Clallam County: Forks, 6. Clark County: Vancouver, 4. Cowlitz County: Silverlake, 2. King County: Enumclaw, 1; Red Mountain, 1. Kittitas County: Cle Elum, 1. Pierce County: 1; Long Mire, 3; Mount Rainier, 461. San Juan Islands, 1. Skamania County: Little Huckleberry Mountain, 1. Snohomish County: Arlington, 3; Soda Springs, 1; Sultan, 1. Whatcom County: Mount Baker, 9. Yakima County: Naches Pass, 4. Localities of unknown counties: Chinook Pass, 8; Greenwater, 2; Lake Cushman, 3; Mora, 2; Mount Adams, 13; Pilchuck Mountain, 1; Stillguamish, 2; Verlot, 1.
     Cicindela depressula eureka Fall. United States.   CALIFORNIA: Humbolt County: Alton, 2; Arcata , Mad River, 39; Blue Lake, 4; Bridgeville near Van Duzen River, 19; Fortuna, 1; Orick, Redwood Creek, 12; Scotia, 1; Van Duzen River, 27. Del Norte County: Requa, 3; Terwah, 1.   OREGON: Benton County: Corvallis, 1. Linn County: Albany, 1. WASHINGTON: King County: Seattle, 1. Whatcom County: Naches Pass, 1.

HYBRIDIZATION BETWEEN *C. OREGONA* AND
C. DUODECIMGUTTATA

Introduction

Cicindela duodecimguttata ranges across most of eastern and central North America from Texas to northern Canada, and from the eastern slopes of the Rocky Mountains to the Atlantic (fig. 17). *Cicindela oregona* occupies regions in and west of the Rocky Mountains to the Pacific coast from southern United States to Alaska (fig. 18).

During the glacial stages of the Pleistocene the two forms were probably isolated from one another, *oregona* to the west of the Rockies and *duodecimguttata* to the east of that mountain range. Since Pleistocene times their ranges have expanded and have come together forming a zone of intergradation on the eastern slopes of the Rocky Mountains, that extends from Colorado to northwestern Canada. This hybrid zone is approximately 50 miles wide in the North Saskatchewan River valley in western Alberta, but it is nearly 1,000 miles wide in northwestern Canada. As far as is known hybridization between these two species occurs in all areas of sympatry.

This study is based on 19 population samples comprising 1,731 adult specimens of which 1,291 were collected in Alberta, 205 in the Northwest Territories, 75 in British Columbia, 70 in Montana, 61 in Saskatchewan, 27 in Alaska and 3 in Colorado (see fig. 34). Additional material from areas east and west of the hybrid zone was obtained from various North American institutions and has been analyzed in the *oregona* and *duodecimguttata* taxonomic sections.

Adult male and female *duodecimguttata* are alike in color, color pattern, and distribution of hair on the head. Hairs cover the frons, top of the
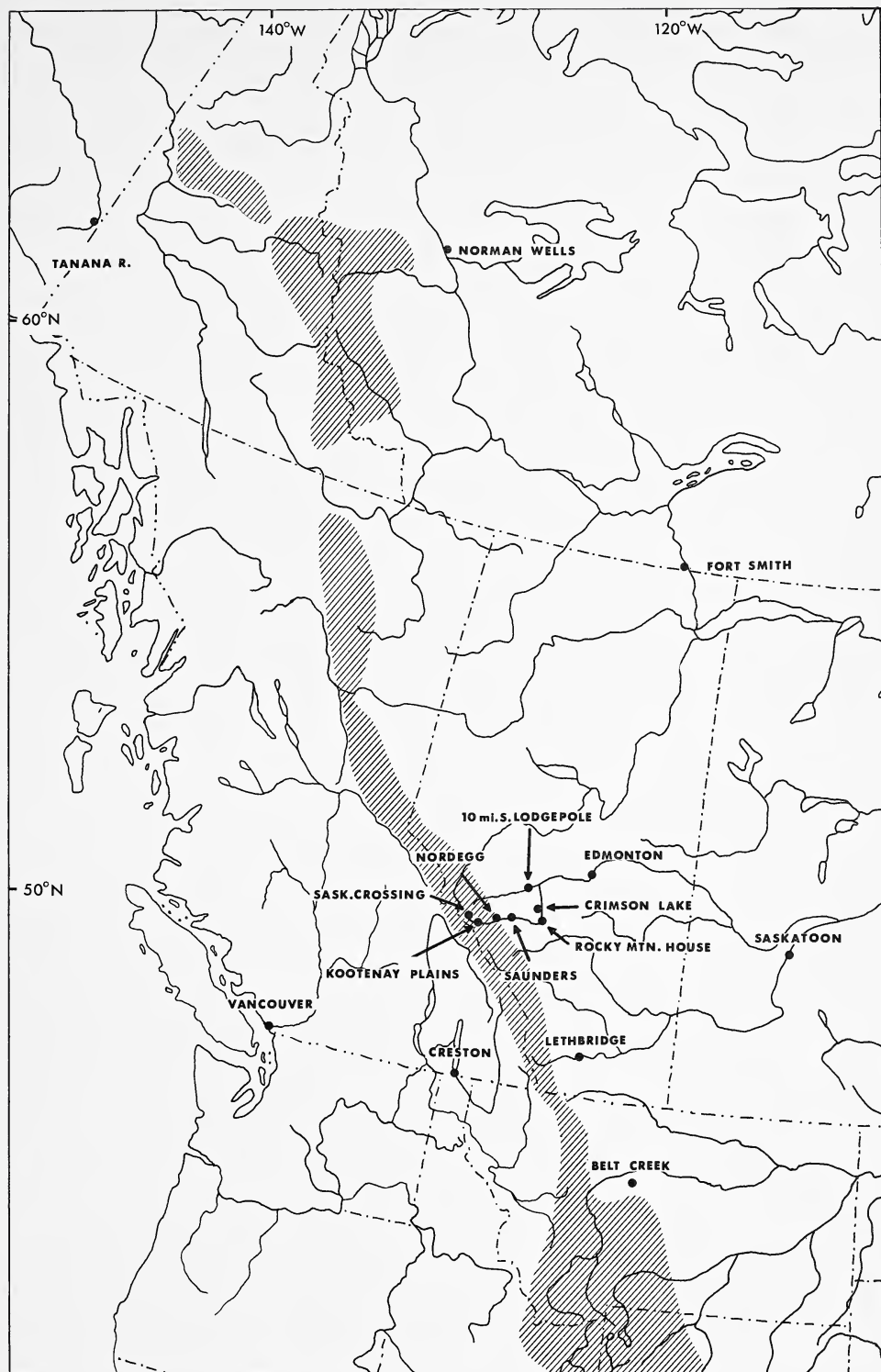
Fig. 34. Locality map of population samples analyzed by hybrid index method. Dark areas indicate elevations above 5,000 feet.

head and post genae. In the western portions of the range of this species individuals are brown dorsally and metallic blue-green ventrally, the prothoracic pleura are coppery, and they have complete elytral patterns (for details see p. 102).

The species *duodecimguttata* hybridizes with the subspecies *oregona guttifera*. Both sexes of this subspecies have similar external features. Hairs are not present on the post genae, frons or top of the head, but a few occupy a small area near the front inner edges of the eyes. The color is identical to that of *duodecimguttata* - brown dorsally and blue-green ventrally, and coppery prothoracic pleura. A reduced elytral pattern is characteristic of *oregona* with humeral and apical lunules each represented by two dots, and the marginal band absent.

Elytral pattern, and distribution of hair on the head were used to develop a hybrid index for *duodecimguttata* and *oregona*. Male genitalia were not used because it seemed preferable to use characteristics occurring in both sexes. High values were assigned to the characteristics of *duodecimguttata*, low to those of *oregona*. Intermediate expressions of these characteristics were scored with intermediate values. These characteristics are illustrated in figs. 11 to 16 and details of assignment of values are given in table 9. In population samples of non-hybrid *duodecimguttata* from western localities, index values range from 4 to 7; and in non-hybrid populations of *oregona*, values range from 0 to 1.

TABLE 9 - Values assigned to diagnostic characters of *C. duodecimguttata* and *C. oregona* used in determination of compound character indices.

Elytral markings and areas of head	Values		
	0	1	2
Humeral lunule	two dots	broken	complete
Marginal band	absent	broken or trace	complete
Apical lunule	two dots	complete	-
Frons	glabrous	hairy	-
Post gena	glabrous	hairy	-

Characteristics of *oregona* and *duodecimguttata* occur in many recombinations in the hybrids. Many specimens are like one of the parental types except for one character while others have index values of 2 or 3. Specimens that have hairs on the frons and head often have hairy post genae. This could be a pleiotropic effect of a single gene, but since the association is inconstant the post genae and frons are treated as separate characters.

Variation in Space

A hybrid index value was determined for each of the 1,731 specimens. A histogram shows the percentage of specimens per index value for each population sample (figs 35-44). The localities from which population samples were collected are illustrated in fig. 34 except those that are represented by only a few specimens.
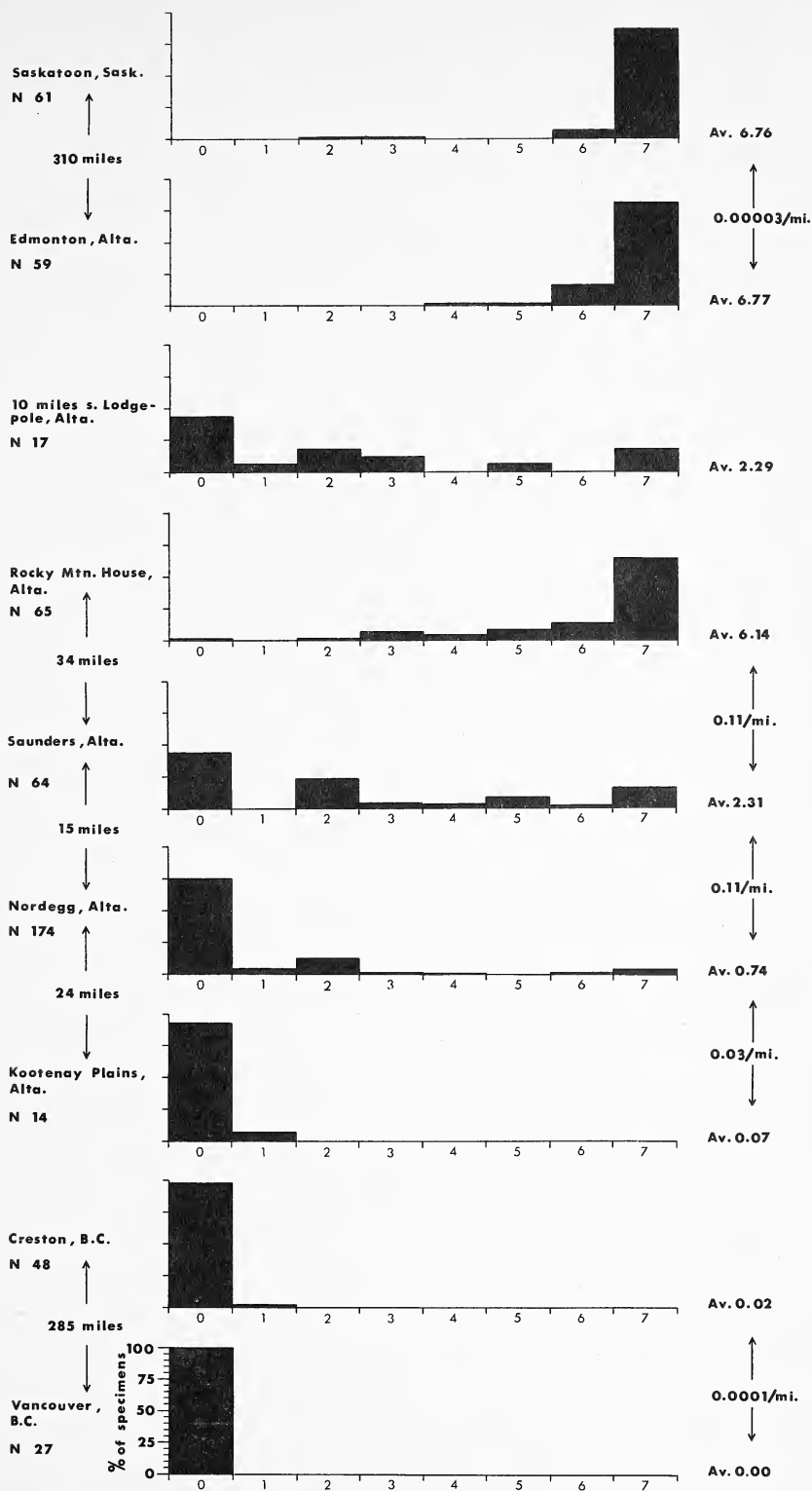
Fig. 35. Changes in the frequency distribution of hybrid index values in population samples of *C. duodecimguttata* and *C. oregona* between Saskatoon and Vancouver. Average hybrid indices and the change in hybrid index per mile on the right, number of specimens and air miles between localities on the left.

cussion; a transect from Vancouver, British Columbia to Saskatoon, Saskatchewan, via the Saskatchewan River System; the Belt Creek, Montana sample; the Boulder, Colorado sample; and a transect from Tanana River, Alaska to Fort Smith, Northwest Territories through Norman Wells, Northwest Territories.

Vancouver —Saskatchewan River Drainage Transect

  In figure 35 three portions of the transect serve to illustrate spatial phenotypic changes between uncontaminated populations of *oregona* and uncontaminated *duodecimguttata* through a zone of intergradation centered in the upper regions of the North Saskatchewan River valley. The transect lies on a west-east plane with the geographically extreme points Vancouver in the west and Saskatoon in the east. The first part includes population samples from Vancouver and Creston. The second portion of the transect is along the western regions of the North Saskatchewan River valley and includes population samples from Kootenay Plains, Nordegg, Saunders, Rocky Mountain House, and Lodgepole. The Edmonton and Saskatoon samples constitute the eastern portion of the transect. Air mile distances and index changes per mile between localities are also given in the figure. Histograms illustrating variation in population samples from Lethbridge, Saskatchewan Crossing, Garth, Crimson Lake, and eight miles south of Lodgepole provide additional data.

  Specimens collected in Vancouver, British Columbia on a sand bank bordering an inlet of the Pacific Ocean all score 0 indicating a pure *oregona* population.

  The sample collected at Creston, 285 air miles east of Vancouver, is composed of 48 specimens of which 21 were taken by Stace-Smith in 1945 and 27 by Ball in 1957. Only one specimen scores 1, while the remaining score 0. Average index change per mile, from Vancouver to Creston is 0.0001 which is negligible and can be attributed to natural variation in the Creston population.

  In 1962 specimens were collected near the Kootenay Plains, 20 miles down stream from the junction of the Banff-Jasper highway and the North Saskatchewan River. River banks are steep near the Plains and are covered with undergrowth to the edge. These banks are therefore not suitable for riparian tiger beetles such as *oregona*. However, many islands formed by sand and clay deposits occupy the river bed and divide the river into several large streams. Most of these islands are sparsely covered with grass and shrubs, and river debris such as drift wood is plentiful. The islands proved to be suitable habitat for *oregona* and another riparian species, *C. repanda*. One specimen scores 1, the other 13 all score 0. The average index value for the sample is 0.07.

  Sand and mud islands that characterize the North Saskatchewan River near Kootenay Plains are also present 24 miles down stream at the Forestry Trunk Road crossing near Nordegg. Two islands divide the river into three large streams. Three road bridges link the islands with one another and the river banks. The southern island bears large shrubs which occupy higher central portions, while grass is abundant throughout. Much of the west half of the island is a clay flat that is periodically covered with water when the river rises. The soil there is basic with a pH of 8.2. There is little organic matter in the soil but

much free lime. Riparian tiger beetles are abundant on the island esp-
ecially along the clay flat. A series of 174 specimens was collected in
1963;the population sample is variable, composed of *oregona, duodecimguttata,*
and hybrid individuals. The index values range from 0 to 7, with the
average at 0.74.   Of the population sample 76 per cent score 0,
four per cent score 7, and 26 per cent have intermediate values. The
average index value increases 0.03 units per air mile from Kootenay
Plains to Nordegg, which is 300 times greater than that from Vancouver
to Creston.

　　　Fifteen air miles east of Nordegg near Saunders the North
Saskatchewan River is narrow and there are no islands. The north shore
is a sandy strip several to 10 feet wide and littered with drift wood.
The river banks descend sharply to the sandy shore line, and are covered
with undergrowth for the most part but grassy clearings are present.
These clearings are probably periodically inhabited by riparian tiger
beetles when the river is high and covers the sandy shore margins.
C. oregona , *duodecimguttata* and hybrid individuals are abundant on the b e a c h
at Saunders from which a series consisting of 64 specimens was taken
in 1963.

　　　The range in index values is 0 to 7 with the average at 2.31. This
is a mean index increase of 0.11 per mile from Nordegg to Saunders,
that is, more than three times the average index increase per mile from
Kootenay Plains to Nordegg. Specimens with an index of 0 comprise
43.8 per cent of the population sample, while those which score 7 con-
stitute 15.6 per cent. Individuals which score 2 to 6 inclusive make up
40.6 per cent of the sample. No specimen has an index value of 1.

　　　Thirty-four miles east of Saunders, at the Highway 11 bridge
near Rocky Mountain House, the river banks are flatter and broader
than those upstream. A mixture of clay, sand and loose gravel forms
the south bank near the bridge. Open patches on which tiger beetles are
active are common.

　　　The index values range from 0 to 7 and the average is 6.14.
Specimens scoring 7 comprise 64.7 per cent of the population sample
while individuals with a value of 0 constitute 1.5 per cent. Most
hybrids resemble *duodecimguttata* more closely than *oregona*. Although non-
hybrid *oregona* is scarce in this predominantly *duodecimguttata* p o p u l a t i o n
many hybrid individuals are present.

　　　Ten miles south of Lodgepole large sand banks flank the north
side of the Brazeau River adjacent to the Brazeau power house. Because
a dam has been erected further upstream, only a little water is present
near the power house and much of the river bed is exposed. On August
12, 1963 a population sample was collected on the sand banks near the
Brazeau power house.

　　　The specimens have index values that range from 0 to 7 but none
score 4 or 6. The average index is 2.29. Because the Brazeau power
house is located near the North Saskatchewan River between Rocky Mount-
ain House and Edmonton, it would appear that the mean index value of a
population sample from that area should fall between 6.14 and 6.77, the
average indices of Rocky Mountain House and Edmonton respectively.

In fact, the average index is approximately 4 less than expected. This may be in part because the Brazeau power house locality although downstream from Rocky Mountain House is 17 miles west of it. In addition, *oregona* and *duodecimguttata* exhibit different habitat preferences (see locality eight miles south of Lodgepole). The pure sand bank near the power house is particularly suitable for *oregona* and harbors a population with a lower average index value than that at Rocky Mountain House.

A sample was collected in 1961 near the Groat Bridge in Edmonton. The average index value is 6.77 and the range is 4 to 7 inclusive. This may be the result of hybridization with *oregona* but is more likely natural variation.

The mean index value of the Saskatoon sample is 6.76 which is a change of 0.00003 index units per mile from Edmonton to Saskatoon a distance of 310 air miles. A small percentage of the sample with values of 2 and 3 is interpreted as natural variation in a non-hybrid *duodecimguttata* population. It is the result of breakdown of the elytral markings, which is probably caused by *duodecimguttata* genes infiltrating from the east, rather than *oregona* genes from the west.

Additional localities in western Alberta

Ten specimens were collected near the junction of the North Saskatchewan River and Banff-Jasper Highway about 20 miles west of the Kootenay Plains. All the individuals score 0 (fig. 36).

Garth is approximately three miles upstream from Rocky Mountain House. The river on the north side is a clear stretch of sand and clay. In May, 1963 a sample was taken on this beach. The index values range from 0 to 7. Most specimens score high and the average index is 5.25 (fig. 37). If this is compared with the histogram for the Rocky Mountain House sample (fig. 38), that was collected on the same day, the mean index difference is 1.24.

Crimson Lake is about nine miles north of Rocky Mountain House. At the east end the water front is sandy. For about 500 feet and at the south end of this beach the sand is light in color and loose and *C. repanda* is abundant. Toward the west end of the beach the ground is a hard and dark mixture of sand and clay and is flanked by a marshy area. Many *duodecimguttata* individuals were active in this area of the beach and *repanda* was absent. Samples were collected at Crimson Lake in 1961, 1962, and 1963. The range of average index values is 6.14 to 6.55 (fig. 39). Several *oregona* specimens were collected here but they are rare. The population is mainly *duodecimguttata* with evidence of *oregona* gene infiltration.

Between Lodgepole and the Brazeau power house many ponds are scattered beside the gravel highway. These are water-filled gravel pits that were dug out for road construction. The soil around the ponds is normally hardened clay covered with grass. The same day the Brazeau power house specimens were taken, I also collected 17 specimens near one of these roadside ponds eight miles south of Lodgepole. The hybrid index values of the roadside samples range from 3 to 7, and the average index is 6.36 (fig. 40). This index average is 4.07 more than the mean index of the Brazeau power house population.

The marked average index difference   between populations of the
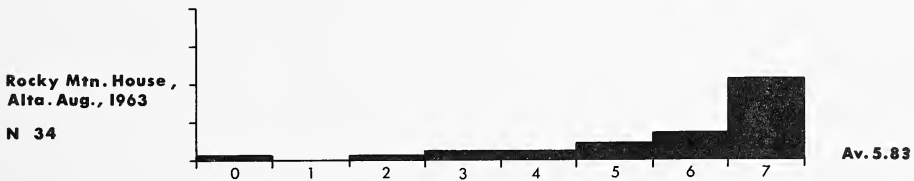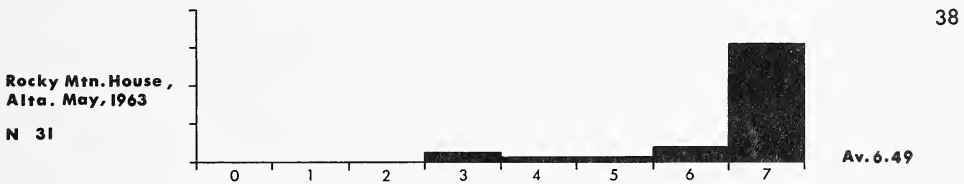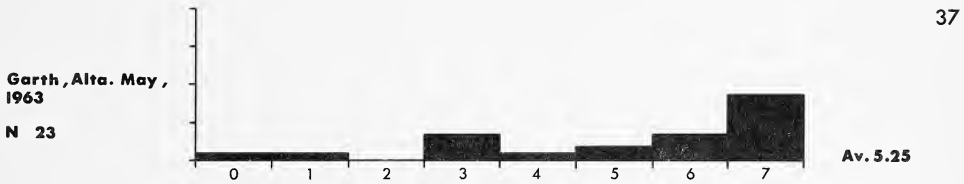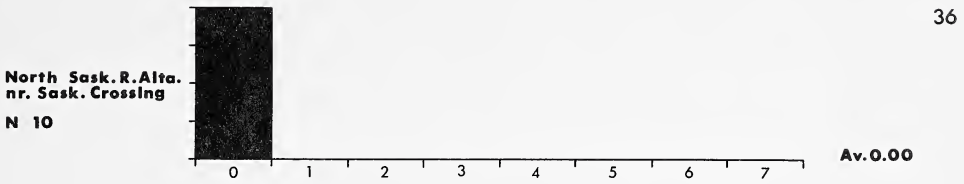
Figs 36-38. Frequency distribution of hybrid index values in population samples of, 36, *C. oregona* from Saskatchewan Crossing, Alberta; 37, *C. oregona* X *C. duodecimguttata* from Crimson Lake, Alberta. N. = no. of specimens.

Brazeau power house and roadside ponds is indicative of different habitat preferences of the two species. Populations of *oregona* are best adapted to pure sand and clean gravel. commonly found along mountain streams. The species *duodecimguttata* normally inhabits edges of lakes, sloughs, and rivers where there are usually flat clearings of dark sand, clay or mud. Wherever these two habitats are available together, at least near the North Saskatchewan River, *duodecimguttata* and *oregona* hybridize. Because the soils around the roadside ponds between Lodgepole and Brazeau power house are predominantly clay, they were probably first inhabited by *duodecimguttata* which invaded these ponds from sloughs etc. nearby. However the effect of *oregona* is evident in these roadside populations.

The Lethbridge sample consists of 310 specimens that were

39

Crimson Lake, Alta.
1961

N 36

Av. 6.50

Crimson Lake, Alta.
1962

N 65

Av. 6.53

Crimson Lake, Alta.
May, 1963

N 33

Av. 6.55

Crimson Lake, Alta.
July, 1963

N 38

Av. 6.43

Crimson Lake, Alta.
August, 1963

N 45

Av. 6.14
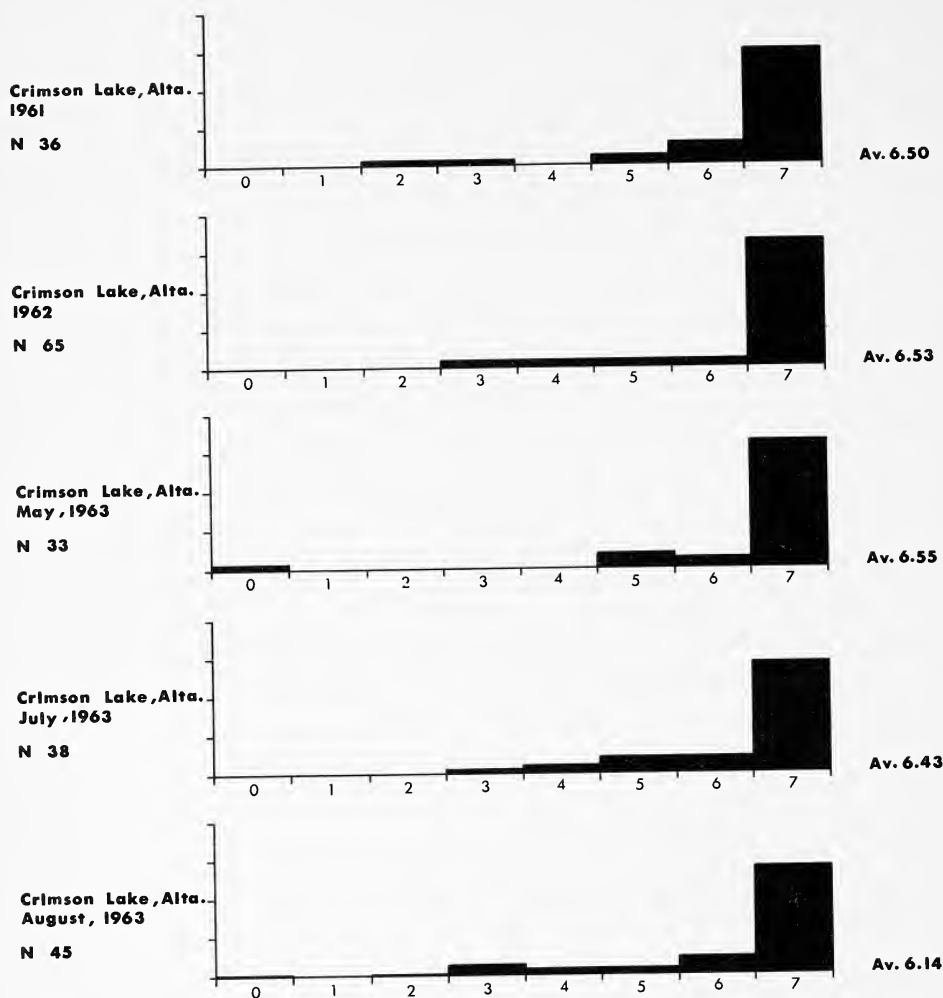
Fig. 39. Frequency distribution of hybrid index values in five population samples of *C. oregona* X *C. duodecimguttata* from Crimson Lake, Alberta. N. = no. of specimens.

collected throughout the summer of 1962 (fig. 41). More than 84 per cent of the specimens are pure *duodecimguttata*. No individuals score 0 or 1. The index range is 2 to 7 inclusive with the average at 6.7. The histogram indicates that some introgression from *oregona* is influencing this predominantly *duodecimguttata* population. In 1960 two *oregona* specimens were collected near Lethbridge. The occurrence of these specimens supports the supposition that variation in Lethbridge is the result of hybridization between *oregona* and *duodecimguttata*.

High River is 30 miles south of Calgary. Of the four representative specimens there are two hybrids, one *oregona*, and one *duodecimguttata*.

In 1925 F. S. Carr collected eleven specimens from Happy
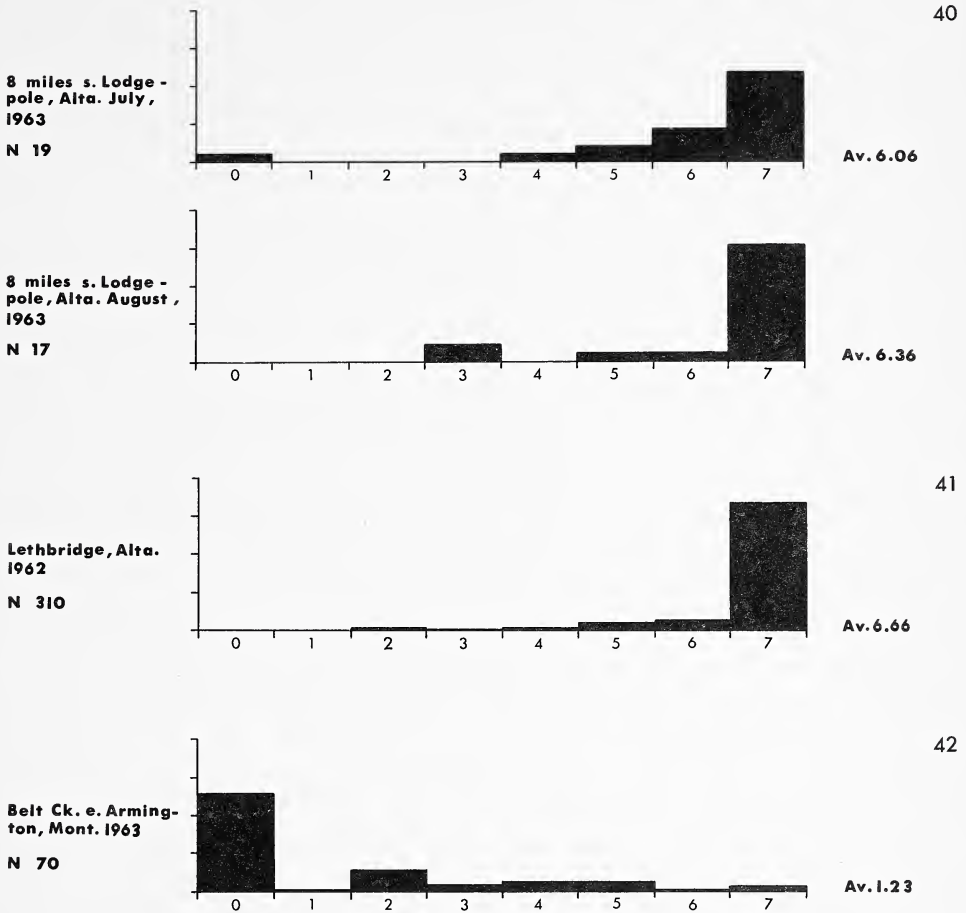
Figs 40-42. Frequency distribution of hybrid index values in population samples of, 40, *C. oregona* X *C. duodecimguttata* from 8 mi. south of Lodgepole, Alberta (2 samples); 41, *C. oregona* X *C. duodecimguttata* from Lethbridge, Alberta; 42,   X *C. duodecimguttata* from Belt Creek near Armington, Montana. N. = no. of specimens.

Valley which is near the Bow River approximately four miles west of Calgary. The sample consists of six *oregona*, two *duodecimguttata* and three hybrids.

Three specimens were taken in 1961 by Wu near Ricinus along the Clearwater River, 20 miles south of Rocky Mountain House. Two individuals are *duodecimguttata* and one is a hybrid.

Two specimens of *duodecimguttata* and one hybrid were collected at Beaver Creek, Alberta. The locality and collector of this small sample are both unknown.

Belt Creek, Montana

In 1963 a sample was collected along Belt Creek just east of

Arlington, Montana.   Specimens of both *oregona* and *duodecimguttata* were
taken with hybrids. All of the index values are represented, but *oregona*
specimens represent 64. 3 per cent of the sample   while two indiv-
iduals score 7 and constitute 2. 9 per cent of the series. The index mean
is 1.23 (fig. 42).

         There are mean differences in index values between males and
females.   Values for males do not exceed 5, and the mean index value
is 0.70.   Thirty-four females show a range in values from 0 to 7 and
their average index is 1.79; that is, 1.09 more than the males which total
36. In other samples, the differences between males and females is less.

Boulder, Colorado

         The   southernmost hybrid sample   is   represented b y two
duodecimguttata specimens and one hybrid which were collected four miles
north of Boulder, Colorado in July 1960. A histogram is not provided
for this sample.

The Alaska - Fort Smith Transect

         This transect, composed of population samples collected at the
Tanana River, Alaska, Norman Wells, Northwest Territories, and Fort
Smith, Northwest Territories extends over a range of about 1, 000 miles
(fig. 34).   Index values were determined for all of the specimens and a
histogram is presented for each of the three samples (fig. 43). Included
in the figure are air mile distances and index changes per mile between
the localities.

         In 1958 Ball collected a series of specimens at a junction of the
Tanana River and the Alaska Highway in southeastern Alaska. Specimens
that have an index value of 0 constitute 88. 9 per cent of the sample, while
two individuals, representing 11. 1 per cent each score 1, because both
have hairs on their heads. The average index value is 0. 11.

         Norman Wells, situated near the Mackenzie River, is approxi-
mately 470 miles east - northeast of the Tanana River locality. Index
values range from 0 to 7 with the average at 2. 68. The average index
change per mile from Tanana River to Norman Wells is 0. 00547. The
sample exhibits a great amount of variation and hybrids outnumber the
parental forms. One specimen scores 0 and two members each have
values of 7.   Thus the parental specimens together constitute only 6. 9
per cent of the sample. In contrast individuals that score 2 occur in the
greatest frequency and make up 69. 9 per cent of the sample. It has been
pointed out earlier that natural variation in an uncontaminated *duodecimguttata*
sample may include specimens indexed from 2 to 7. Similarly a pure
oregona population can have individuals that score 1 as well as 0. Thus
a true hybrid is considered to have a value of 2 or 3. Because speci-
mens with scores of 2 and 3 dominate the Norman Wells sample it is
regarded as a predominantly hybrid population sample and is the only
one of its kind in this study.

         A sample was collected at Fort Smith near the Slave River, 580
miles southeast of Norman Wells. Index values range from 2 to 7 with
the average at 5. 97. From Norman Wells to Fort Smith the change per
mile is 0. 00567 units. No specimens have values of 0 or 1. Individuals
that score 2 occur in lowest frequency while those with a value of 7 are

43

Tanana R., Mi.1281
Alas. Hway. Alas.

N 27

470 miles

Norman Wells,
N.W.T.

N 43

580 miles

Fort Smith, N.W.T.

N 162

Av. 0.11

0.00547/mi.

Av. 2.68

0.00567/mi.

Av. 5.97

Fig. 43. Frequency distribution of hybrid index values in population samples of *C. oregona* X *C. duodecimguttata* from Alaska and the Northwest Territories. Average hybrid indices and the change in hybrid index per mile on the right, number of specimens and air miles between localities on the left.

44

Nordegg, Alta. 1961

N 83

Av. 2.60

Nordegg, Alta. 1962

N 219

Av. 0.97

Nordegg, Alta. May,
1963

N 62

Av. 0.98

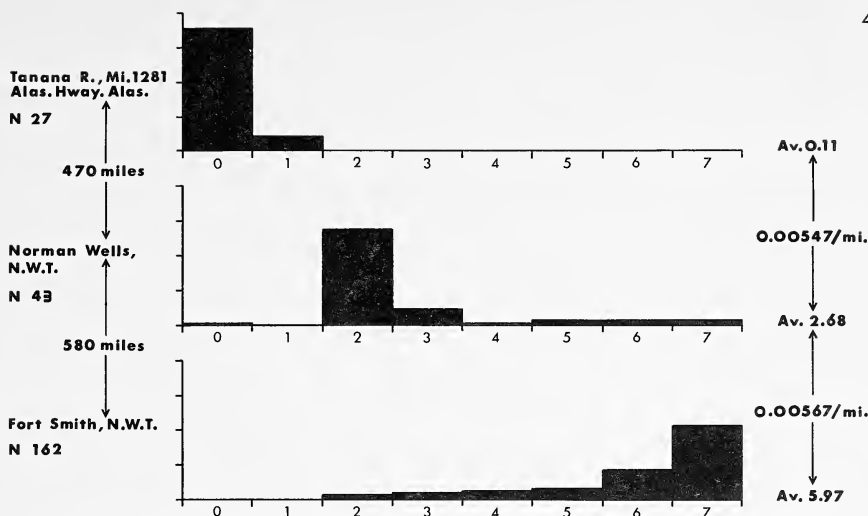Nordegg, Alta. July,
1963

N 54
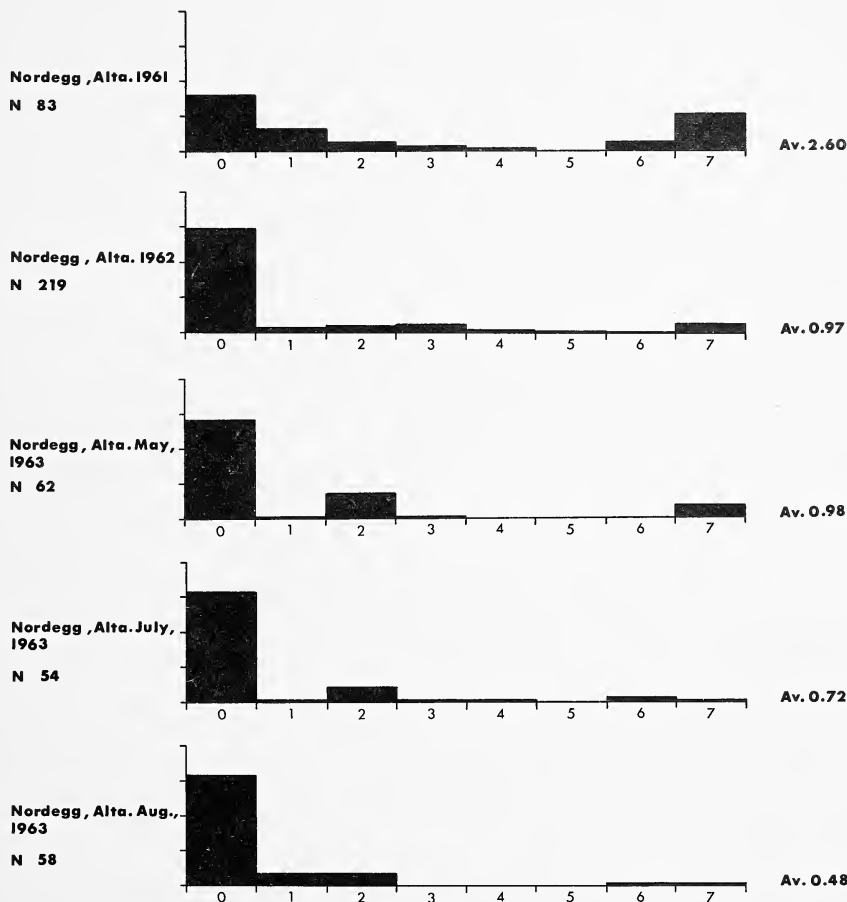
Av. 0.72

Nordegg, Alta. Aug.,
1963

N 58

Av. 0.48

Fig. 44. Frequency distribution of hybrid index values in population samples of *C. oregona* X *C. duodecimguttata* from Nordegg, Alberta. N. = no. of specimens.

most numerous.   The number of specimens increases with increasing index values.   The sample is therefore a *duodecimguttata* one affected by introgression of *oregona* characteristics.

Of the five specimens taken at Canol near the Mackenzie River opposite Norman Wells there are one *oregona,* one *duodecimguttata* and three hybrids.

One *oregona* specimen and one hybrid were taken at Fort Simpson which is 290 miles southeast of Norman Wells near the mouth of the Liard River.

Variation in Time

Annual and seasonal variation in index values appear to be typical of most localities in the western section of the North Saskatchewan River. Variation is illustrated by histograms for population samples collected in the Nordegg area, near Rocky Mountain House, at Crimson Lake, and in an area eight miles south of Lodgepole.

Nordegg

Evidence of extensive hybridization is clearly shown in the histogram for the 1961 sample (fig. 44). Index values range from 0 to 7 and the average is 2.60. Specimens of *oregona* are most common, constituting 39.8 per cent of the sample. Specimens of *duodecimguttata* follow in number and are 25.3 per cent of the sample. The remaining portion of the series is formed by intermediate specimens which are mainly at the low end of the index scale.

The situation is markedly changed in 1962. A shift toward the low end of the histogram occurs. Specimens that score 0 increase to 74.2 per cent. while specimens with an index of 7 drop to 6.8 per cent of the sample. The average index is 0.97, a drop of 1.63 from the previous year. There is a further depletion in *duodecimguttata* numbers in 1963 but there is very little general change in the frequency of indices from that of 1962.

This may be a phase in fluctuating populations of the parental forms. However many more years of sampling at Nordegg would provide a clearer picture of annual variation in these populations. Analysis of Nordegg population samples collected in May, July, and August, 1963, revealed a slight trend in decrease of average index values throughout the summer.

Values for the May sample, range from 0 to 7, excepting 4, 5, and 6, with the average at 0.98. The sample is therefore predominantly *oregona* specimens (71 per cent), some hybrids closely resembling *oregona,* and five phenotypic *duodecimguttata* individuals.   In the July sample more classes are present, and only index value 5 is not represented.   The index value is 0.72, which is 0.26 less than that of May.   Specimens with high index values are all but absent from the August sample. Most of the specimens are at the low end of the scale with the largest number at 0.

There also appears to be a seasonal change in the relative numbers of males and females at Nordegg.   The ratio of males to females in the May population sample is approximately 3 to 4 (27 males and 35 females), but males outnumber females in the July sample, 2 to 1 (36 males and

18 females).   In the August series, the ratio of males to females is approximately 3.5 to 1 (45 males to 13 females).

Rocky Mountain House

The range in index values for the May series is 3 to 7 with the average at 6.49. Of the sample, 24 specimens score 7. Thus the population sample is basically a *duocecimguttata* one somewhat contaminated by *oregona* genes (fig. 38). The August sample is more variable and all the index values except 2, are represented. The average index value is 5.83 which is a decrease of 0.66 from the May value.

Relative members of males and females also change seasonally, and parallel the change which occurs at Nordegg. The ratio of males and females in May is 1 to 1 (15 males and 16 females), while in August there are approximately three times as many males as females (26 males and 8 females). This difference however is not statistically significant.

Crimson Lake

The histogram for specimens collected at Crimson Lake in 1961 shows a mainly *duodecimguttata* population (fig. 39). One specimen has an index value of 2 and one has a value of 3. The mean index is 6.50. The range of index values for 1962 is 3 to 7 with the mean value at 6.53. The average index change from 1961 to 1962 - 0.03 - is quite small. In 1963 the mean value decreased by 0.19, and the range in index values is 0 to 7.   The population sample however is largely a *duodecimguttata* one much like those of the two previous years. The major difference is that *oregona* specimens are present in the 1963 series, but they are rare.

From May to August a general decrease occurred in the mean index values of the Crimson Lake samples. This seasonal index change corresponds with that of Nordegg and Rocky Mountain House.

Ten males and 23 females in May, 21 males and 17 females in July, and 25 males and 20 females in August were collected   in 1963. The sex ratio is two females to one male in the May sample, but is one to one for July and August.

Lodgepole - eight miles south

In 1963, small series of 19 and 17 specimens were collected at this locality in July and August respectively. Seasonal differences between the two samples do not coincide with those of Nordegg, Crimson Lake, and Rocky Mountain House but the samples are probably too small to indicate real differences. However there seems to be a shift from a lower average index value earlier in the season to a higher average value later in the season.   Both samples consist mainly of *duodecimguttata* specimens but with a few hybrids (fig. 40).

The males and females are present in about equal numbers in both population samples, with nine females and 10 males taken in July, and nine females and eight males collected in August.

Notes on mating

During the summer of 1962, in the Nordegg area, 26 pairs of mating tiger beetles were collected. This is a phenotypically varied

group, including both parental species and hybrids. Hybrid indices were determined for the specimens. Then, a chi square test for independence was applied to find out if specimens of a particular index more often selected a mate of the same index value. It failed to show any selection in mating.

On several occasions I have taken *repanda* in copulation with *oregona* and also with *duodecimguttata*, but no hybrids have been found. It is doubtful that gene exchange takes place between *repanda* and *oregona* or *duodecimguttata*, to the extent that it does between *oregona* and *duodecimguttata*.

Discussion

The kind of hybridization between *oregona* and *duodecimguttata* can be classified as one of introgression (Anderson 1949), and secondary intergradation (Mayr 1942). Introgression, as described by Anderson, is the incorporation of genes of one species into the gene complex of another species. Mayr states that secondary intergradation has occurred when "Two units, now connected by a steeply sloping character gradient were separated completely at one time and have now come into contact again after a number of differences have evolved." Mayr (1963) regards the species involved in this kind of hybridization as semispecies in that they show some of the characteristics of a species and some of subspecies. Many such cases have been described for birds, mammals, fish, amphibians and some invertebrates. These are too numerous to mention here but many are cited by Dobzhansky (1951), Mayr (1942, 1963), Mecham (1961), and Sibley (1961).

The situation in western Alberta and northwestern Canada described above seems to be the result of secondary contact between the formerly isolated vicariant species *oregona* and *duodecimguttata*. Their phenotypic differences probably arose under different selective forces acting on allopatric populations. Breakdown of external barriers between them allowed their ranges to expand and come into contact. This has resulted in hybridization. Speciation of *oregona* and *duodecimguttata* was probably initiated in early Pleistocene times, but the process has not been completed. Climatic changes during the Pleistocene undoubtedly had a profound influence on the distribution of these two forms as they did on other North American animal species (see Blair 1951). Unlike the vertebrates, however, neither locations of refugia for these tiger beetles nor their population movements during the Pleistocene are known because of the lack of a fossil record. The history of this zone of secondary intergradation is therefore speculative, and is based on the present distribution of both species, and current concepts of events during the Pleistocene. During the early period of *oregona* subspeciation, populations of *duodecimguttata* were not present in western regions they now occupy. Shortly after the development of *o. guttifera,* perhaps *duodecimguttata* reinvaded western Canada east of the Rocky Mountains. Because, at the present time, few southern populations of *duodecimguttata* reach the eastern front of the Rockies in Colorado and New Mexico, the present western limits are presumed to be the extent of the western limits of *duodecimguttata* during the late Prairie interglacial. If any hybridization did occur in Prairie times it took place where the two species are presently sympatric. However, any evidence of pre-Recent introgression would be masked by the present hybrid belt. Hybridization probably did occur in southern

regions during the Wisconsin glaciation since no indication of introgression is evident in southern populations.

The hybrid belt between *oregona* and *duodecimguttata* is widest in northwestern Canada and narrowest in western Alberta. Individual specimens of *oregona* and *duodecimguttata* exist in all areas of greatest variability including the Norman Wells population where they are outnumbered by intermediates. Width of the zone of intergradation is recognized as spatial limits of extreme variation.

There does not seem to be any reduced viability or fertility in the hybrid tiger beetles and they are present in large numbers in the Norman Wells sample described above. A composite of isolating mechanisms, although hardly pronounced in southern populations of *oregona* and *duodecimguttata*, may have become more completely developed than in northern populations of the two species before they made contact. This may account in part for the varying width of the zone of intergradation between *oregona* and *duodecimguttata*.

Somewhat analogous is the intergradation zone of the European crows *Corvus corone cornix* and *C. c. corone* (Mayr 1942, pp. 265-266), and that of the North American grackles *Quiscalus quiscula quiscula* and *Q. q. versicolor* (Huntington 1952). Dobzhansky (1951) attempts to explain the irregular width of the intergradation belt of the crows. He suggests that oldest regions of the zone are narrowest where isolating mechanisms have had more time to become established. Mayr (1942) does not believe this explanation corresponds with the facts presented by Meise, who observed the width of the hybrid zone of *Corvus* is determined by local ecological factors. Further, narrow stretches of intergradation occur in both recent and older parts of the zone. Because in *Quiscalus*, Huntington (1952) observed no reduced viability or fertility in the intermediates, he feels Dobzhansky's explanation is inadequate in principle, and suggests that increased mixing due to migration, and selective forces favouring the intermediate in a rapidly changing environment are the two main factors affecting the width of area of intergradation.

Because the width of intergradation zones is determined largely by isolating mechanisms, it is appropriate to discuss variation in the width of the tiger beetle hybrid zone in the light of two sets of theories on the origin of isolating mechanisms.

For several hypotheses natural selection is believed to be responsible for the promotion of isolating mechanisms. One representative hypothesis postulates that intermediates are of lowered fertility or viability compared to parental forms. From this it is argued that individuals which enter into mixed pairs will eventually be eliminated from both populations because the hybrids they produce are being selected against. In time, as isolating mechanisms are perfected, the zone of intergradation is contracted. This is essentially Dobzhansky's view.

A second hypothesis treats the origin of isolating mechanisms as an incidental by-product of genetic divergence in isolated populations (Muller, 1940) rather than as the direct result of selection for reproductive isolation. Mayr (1963) points out that many isolating mechanisms vary geographically.

Because many isolating mechanisms have ecological components, any changes in incipient species are certain to affect their isolating mechanisms. The narrowness of the zone in western Alberta can be

due in part to different habitat preferences (see p. 157). Clay, or mud, or sand with organic material, seem to be preferred by *duodecimguttata*, while soils of pure sand or clean gravel are typical *oregona* habitat. In the north, where the intergradation zone is wider, both species may be more broadly adapted. The broader northern zone may also be an effect of better adaptation of intermediates to the northern environment than to that of the south. However, in order to understand this zone of intergradation more completely, ecological investigations should be undertaken.

The elytral pattern of *duodecimguttata* is complete in western parts of the species range but it is reduced in eastern and southern regions. The full pattern also appear in the zone of intergradation. Eastern *duodecimguttata* specimens often have *oregona*-like elytral markings (see p. 102). This situation may be interpreted as sympatric character divergence, which may be described as follows. Two closely related species of animals overlap geographically. Their differences are emphasized in areas of sympatry so that both species are easily recognized. In ranges where one of the species occurs alone it closely resembles the other species.

For several reasons it is doubtful that the variation in the elytral pattern of *duodecimguttata* is evidence of character displacement. Some workers observe that character displacement occurs within regions of overlap (Brown and Wilson 1956, Mayr 1963). The complete elytral pattern of *duodecimguttata* is present in the hybrid belt in western Alberta but it is also characteristic of populations far beyond the zone of overlap (fig. 17). In addition variation in elytral pattern of *duodecimguttata* is not complemented by similar clines of other characters. For example the shape of the median lobe of the male is uniform throughout the range of *duodecimguttata* except in the hybrid zone where there are many intermediate shapes ranging from that of *oregona* to that of *duodecimguttata* (see p. 97). Similarly, hairs are present on the frons, top of the head, and post genae of *duodecimguttata* throughout the species range except in the area of intergradation. Furthermore, since there is random interspecific mating in the zone of hybridization, the difference in markings does not serve as an isolating mechanism.

PHYLOGENY AND ZOOGEOGRAPHY OF THE NORTH AMERICAN
SPECIES OF THE *MARITIMA* GROUP

Phylogeny

The ancestral form of the North American species of the *maritima* group is necessarily reconstructed from features that are widespread among extant species because no fossils are available. The rationale and principles used in re-constructing the characters of a hypothetical ancestor are explained in Cain and Harrison (1960). The characters of the ancestral species were probably as follows: dorsum, brown, opalescent; venter, metallic blue-green; thoracic pleura, copper colored; humeral, apical, and middle lunules, and marginal band, complete;

lunules narrowly expanded as shown by *hirticollis* or *repanda*; shapes of the individual markings like those of *repanda*; hairs present on the head between the eyes; features of the male genitalia as they are now; flanges of the median lobe comparatively narrow like those of *hirticollis* or *repanda*; fields *a*, *b*, and *c* of the internal sac lightly aculeate; sclerites 1, 2, 3, 4, and 6 of the internal sac general size and shape of extent species; sclerite 5 large like those of *hirticollis* and *repanda*; sclerite between 2 and 6 intermediate size between that of *hirticollis* and *oregona*; pronotum of the larva densely pilose. The species was a riparian form, and it gave rise to three lineages (fig. 45).

The first derivative stock (1) was perhaps characterized by an alteration of the elytral pattern in which the posterior portion of the humeral lunule was produced obliquely towards the median line; within the male's internal sac, field *a* and sclerites probably became respectively densely aculeate and considerably reduced; the pronotum of third instar larvae was probably quite pilose.

This primary stock ultimately gave rise to the species *limbata*, *bellissima*, *theatina*, and *columbica*. The species *limbata* and *bellissima* appear to be most closely allied. The subspecies *limbata hyperborea*, and *bellissima* each have the posterior tip of the humeral lunule extending almost to the middle band; sclerite 5 of the male internal sac has been lost in these species; the riparian habit was abandoned and both species are sand dune inhabitants. Beside the differences in shape of their median lobes, *limbata* is very hairy between the eyes and *bellissima* is less so. In this respect, *bellissima* has departed further from the ancestral stock than has *limbata*. The southern races of the latter species, however, have lost almost all of the dark pigment of the elytra.

The species *theatina* and *columbica* appear to be more closely related to each other than they are to the other two. A humeral lunule whose posterior tip is briefly extended is a diagnostic feature of *theatina* and *columbica*; a very large triangular sclerite has evolved between 2 and 6 of the male internal sac, and sclerite 5 has not completely disappeared. They differ mainly in two characters: *theatina* is densely hairy between the eyes and lives on sand dunes, while *columbica* is sparsely hairy between the eyes and has retained the riparian ancestral characteristic.

The proposed course for *limbata*, *bellissima*, *theatina*, and *columbica* is not presented in a dichotomous scheme in fig. 45 because different arrangements can be devised on the basis of other similarities among the four species. Distribution of hairs on the head, condition of elytral pattern, color, or habitat preferences each could be used to erect a different hypothetical course, but each of these would imply a greater amount of parallelism or convergence than is required in the scheme I have suggested.

The second lineage (2) is represented by the species *hirticollis* which is somewhat remotely allied to the other existing North American species of the *maritima* group. This form evolved: a humeral lunule the posterior tip of which is distinctly hook-shaped; a comparatively pronounced twirl in sclerite 4, and a very large sclerite between 2 and 6 of the male internal sac; and it has retained a densely pilose pronotum of the third instar larva.

The third ancestral stock (3) probably developed or retained a c-shaped humeral lunule; field *a* of the male internal sac remained lightly

aculeate and sclerite 5 increased in size.Secondary setae probably sparse-
ly covered the pronotum of the third instar larva;and the species was   most
likely riparian.   This ancestral stock gave rise to the species *repanda*,
depressula, oregona, and *duodecimguttata.*

          The species *repanda* appears to be less closely related to the other
three species than they are to one another. Within the male internal sac,
sclerite 5 has become very well developed and the sclerite between 2 and
6 has been lost; *repanda* ranges across North America but no introgression
is evident with the other three species in areas of sympatry.

          Evolving from a *repanda* like ancestor, the stock which gave rise to
depressula developed a median lobe with broad flanges, lost most of the frontal
hairs, and developed a modified pattern of white elytral markings, which
at first were extensive but subsequently became much reduced. Also the
ancestral brown color of the dorsum was replaced by blue and green in
the stock which developed reduced markings, and in the east the lowlands
were abandoned by this form for life high in the mountains. Simultaneously
the larva of this derivative form lost most of the pronotal hairs charact -
eristic of the pronotum of the ancestral stock.

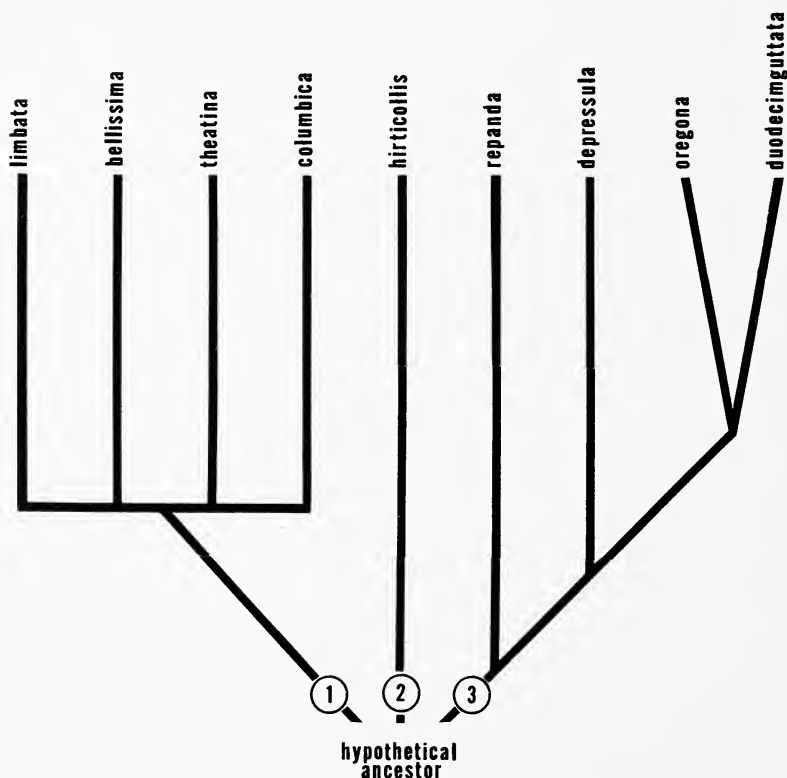   Another derivative stock from a *repanda* - like ancestor, was the

Fig. 45. Hypothetical phylogeny of the North American species of the *martima* group.

progenitor of *duodecimguttata* and *oregona*. This stock developed at first slightly reduced elytral markings. Subsequently it became divided into   two geographically isolated groups the western of which lost the frontal hairs, developed strongly reduced elytral markings, and throughout much of its range the brown color of the dorsum was replaced by green, blue, or purple, and the pleural sclerites became blue or green. The pronotum of the larva gradually lost much of the pubescence evident in the ancestral larva. This western isolate is the species *oregona*. In the eastern isolate, the elytral markings were also reduced, and blue and green color of the dorsum appeared. The mutations producing reduced markings became widespread replacing the ancestral condition throughout most of the range of the species. Hairs on the pronotum of the larva were reduced in number.   This eastern isolate is the species *duodecimguttata*. Following a period of separation too short to permit the development of complete reproductive isolation the eastern and western stocks met one another and a narrow zone of hybridization developed in the area of contact.

This scheme requires postulation of an appreciable amount of parallel evolution. Frontal hairs were lost or reduced four times. Green or blue color of the dorsum was developed six times. Broad flanges on the median lobe were developed six times. The primitive elytral pattern was lost five times, but by two different phyletic branches. In one of these extensive reduction of lateral pigmentation took place. The other type of pattern breakdown was developed by increased pigmentation.

Thus these species, together form a structurally uniform group in which a number of similar structures have arisen independently. This suggests that the group possesses a good degree of evolutionary homo-dynamy (Bock 1963). This principle is defined as follows: "The number of times and ease with which an identical or very similar feature may arise independently within a group depends upon its degree of evolutionary homodynamy which in turn depends upon its common genetical-developmental potential. " In the light of this principle similar structures that have arisen independently in the North American *maritima* group are considered to be homologous in the broad sense, which is defined by Bock as follows: "homologous features (or conditions of the features) in two or more organisms are ones that can be traced back to the same feature (or condition) in a group possessing a high degree of evolutionary homodynamy. "

Zoogeography

The following account of the development of the distribution of the North American species of the *maritima* group is hypothetical. Movements, and times and place of origin of extant species are necessarily constructed on the basis of: distribution and morphological features of the species, geological and climatic events of the Tertiary and Pleistocene in North America, and rates of evolution in some other groups of insects.

Insects generally develop modifications of structural features at a slow rate. Many fossil species of the early or middle Tertiary closely resemble existing species (Linsley 1958, Ross 1958, Becker 1963, Quate 1963, Sabrosky 1963, and Sturtevant 1963). Most of these are members of recent genera. However, Zeuner (Sylvester - Bradley, 1963), by means of analysis of 212 species of fossil Apoidea, Lepidoptera,

and Saltatoria, reckons that excepting the honey-bee all living species evolved in the Pleistocene. He estimates half to one million years is a reasonable time required for the evolution of a full species. Zeuner further notes that no insect species are known with certainty to have survived from the Miocene (see Zeuner 1943, for more information of the time factor in evolution of insects). There is no evidence of recent vigorous evolution within the North American species of the *maritima* group. Indeed these are rather primitive in comparison with other species groups of *Cicindela*. The ancestral stock of the *maritima* group may have been in existence during the early Tertiary. Living species may have evolved during the later Tertiary or early Pleistocene.

Historical events which may have effected geographical isolation and subsequent speciation of tiger beetle populations are of importance. Thus it is necessary to review briefly geological and climatic changes in western North America during the Tertiary and Pleistocene (see Blackwelder 1948, King 1958, MacGinitie 1958, Martin 1958, and Mengel 1964).

The Tertiary was marked by several periods of crustal disturbances. Early Tertiary was a time of extensive mountain building through the west, and it was then the initial Rocky Mountain system was thrust up. Crustal folding was renewed in the middle Tertiary (late Miocene). Gentle folding in the Rockies prevailed. Disturbances were evident in coastal and southeastern California, and southern Nevada, while other mountains were widely distributed throughout the American west. A chain of volcanoes was built up along the east flank of the Sierra Nevada and Cascade Mountains. Large basins were produced, many of which became lake basins. At the close of the Tertiary (late Pliocene) once again crustal folding occurred along the Pacific coast, and in Nevada and Utah. The modern California Coast Range, Wasatch and Ruby Mountains and many others were elevated during this period. The southwestern plateau was raised to its present level, and most of the interior drainage systems were renewed.

Early Miocene and most of the Pliocene were periods of relative quiet. Stream systems wore down western mountains to scattered hills, and extensive plains were formed on which large lakes drained or were filled.

The climate in the early Tertiary was warmer than now. Tropical forests filtered into the north Temperate Zone while temperate conditions prevailed in Rocky Mountain regions. In the Miocene the climate became cooler and temperatures steadily decreased into the Pleistocene. Simultaneously climatic zones moved southward and southwestern regions became drier.

The end of the Tertiary and beginning of the Pleistocene was characterized by the gradual development of mountain glaciers and continental ice masses. There were five major glacial stages in North America, the Nebraskan, Kansan, Illinoian, Iowan, and Wisconsin. Between these occurred long warm periods, the Aftonian, Yarmouth, Sangamon, and Prairie.

In glacial periods glaciers extended southward along mountain ranges. These gave rise to rivers which descended onto open basins where much sand and glacial till was deposited. Large lakes developed

in nearly all western basins.

Climate and vegetation similar to those of the present time were prevalent in interglacial periods in northern latitudes.

All of the North American species of the *maritima* group live in subarctic to warm temperate regions. Perhaps the ranges of *hirticollis* and *oregona* extend for a short distance into Mexico but for the most part they are northern forms. The species, *hirticollis* and *repanda* are almost transcontinental and inhabit regions from the Cascades in the west to the Atlantic coast. Ranging from the Atlantic seaboard to the eastern slopes of the Rockies *duodecimguttata* is the only true eastern form. Inhabiting areas from the Rocky Mountains to the Pacific coast *oregona* is the western counterpart of *duodecimguttata*. The species *depressula* is restricted to high elevations of the Cascade Range and Sierra Nevada, and in river valleys near the Pacific coast from northern California to southern Alaska. The species *limbata* inhabits areas just east of the Continental Divide. Further south, however, populations are found in Kane County, Utah (*l. albissima* Rumpp). The ranges occupied by *bellissima*, *theatina*, and *columbica* are rather restricted: San Luis Valley in south-central Colorado, is the entire range of *theatina; bellissima* occurs on sea beaches in western Oregon and southwestern Washington; while *columbica* exists in southeastern Washington on beaches of the Snake River.

Knowledge of the distribution of the North American species of the *maritima* group supports the premises that: the ancestral species was a cool adapted form, and mountain ranges of western North America are effective geographical barriers particularly the Rocky Mountain system.

The relationships of the Nearctic species of the *maritima* group to those of the Old World members are not understood (but see Papp 1952), so speculation on time and direction of intercontinental movements is not warranted. However it seems certain that such movements did occur, probably by way of a Bering land bridge (see Gressitt 1963). The hypothesis which follows is based on the as yet unestablished premise that all the Nearctic species are more closely related to one another than to any Palearctic species.

The primitive ancestor of the North American species of the *maritima* group may have inhabited cool - temperate: regions of North America in late Miocene. By virtue of its habits it may have filtered southward along alpine river systems near revived mountains of western North America. It may have assumed a reticular distribution among these mountains and in cooler regions further east. By the continuous folding of strata, and volcanic eruptions, populations probably became disjunct and geographically isolated. The first three derivative stocks may have been established during the course of this unsettled period.

Very little can be said about the place of origin and geographical movements of *hirticollis* because of its present vast range and widespread sympatry with *repanda, oregona,* and *duodecimguttata.* It is probably a relatively old form.

The derivative stock that gave rise to *limbata, bellissima, columbica,* and *theatina* , may have ranged throughout cooler regions of western North America up to the late Pliocene. Western North America had been worn down to extensive plains. Mountains were no longer effective geographic barriers, and sandy habitats occurred abundantly near the coast, near

lakes and rivers, and in dry areas remote from water. Perhaps during its existance the ancestral species became more generally adapted and improvements of functions allowed it to inhabit sandy environments in arid regions, but it also continued its riparian habits. The renewed crustal unrest of the later Pliocene probably disbanded and isolated populations, that evolved into *limbata, bellissima,* and *theatina.*

The species *limbata* may have developed as a sand dune inhabitant on the northeastern side of the revived Rocky Mountains in late Pliocene or early Pleistocene. The original form probably resembled the boreal subspecies *l. hyperborea.* Southern populations were probably established during cooler glacial periods. See Rumpp (1961), for some ecology and mechanism of lost of elytral pigmentation in southern populations of *limbata.*

The ancestral stock of *bellissima,* probably became isolated on the Pacific coast by the renewed folding of the Coast Range in late Pliocene or early Pleistocene.

Populations that evolved into *columbica* probably became locked in by the Sierra Nevada and Rocky Mountains perhaps in the early Pleistocene. Within this area they retained the riparian habits of the ancestral stock.

The species *theatina* may have originally been isolated from other related populations to the east of the Continental Divide in Colorado. It perhaps had a greater range than the San Luis Valley to which it is now restricted.

The ancestral stock from which *repanda, depressula, oregona,* and *duodecimguttata* evolved may have originally been isolated to the east of the Rockies. It eventually became transcontinental, probably in early Pliocene.

The place and time of origin, and subsequent geographical distribution of *repanda* is obscured because it ranges throughout most of temperate North America and is sympatric with several related species, and perhaps speciated before late Pliocene.

The species *depressula* may have developed in late Pliocene. Primitive populations of *depressula,* represented by *d. eureka,* on the west side of the Cascade Range and northern Sierra Nevada probably became geographically segregated from the form which gave rise to *duodecimguttata* and *oregona.*

The common ancestor of the species *duodecimguttata* and *oregona* probably occupied the entire cool temperate North America during the middle Pleistocene. The extant species may have been formed during the middle Pleistocene. Dissection of the range of the ancestral stock took place in glacial periods of the later Pleistocene when ice masses covered Canada and glaciers spread southward on high mountain ranges. The species *duodecimguttata* evolved in the east and *oregona* in the west with the Rockies acting as the major geographical barrier. In glacial periods it is doubtful that populations of *duodecimguttata* merged with those of *oregona* in southeastern regions of the Rockies for no evidence of that exists. Hybridization between these species is proof of their close relationship and that their reproductive isolating mechanisms have not yet become fully developed. Perhaps hybridization between them was more extensive in earlier interglacial periods and their isolating mechanisms have become gradually more effective with each successive glacial period.

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Quaestiones

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QUAESTIONES ENTOMOLOGICAE

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Guest Editorial —   **Two Cultures and the Information Explosion**

We live today in a dangerously unstable and incongruous world. As a travelling scientist in recent years I have dined with friends whose principal problems were calories and obesity and hurried through gloomy alleyways where starving children slept on the pavement for want of a better home or shelter. I have been plied with cocktails in foam-padded chairs at near the speed of sound over the Pacific Ocean and photographed foot-weary peasants, miles from their village, overburdened by their precious loads of firewood. All in a world whose population will double by the end of the century; and more than half the present population is undernourished, despite a level of science and technology which could probably solve the problem within a generation.

Has the scientist anything more to offer society than the extra miles per hour, the new antibiotic, the faster computer, or the hydrogen bomb? I feel that he has and he must, but he is handicapped by the weight of his own information explosion and by its effect upon his education and later professional outlook.

I suggest that in teaching and research we are developing science too much as a technical tool and tend to ignore its value as a guide to human thought and relationships. Before the scientist can play a more effective role in society, he must first put his own house in order. He must learn to contain and handle his own information explosion. Surely it is this explosive growth in scientific and technical

knowledge which is the really unique phenomenon in the history of human society. There is abundant evidence that the population explosion is one consequence of the information explosion, although perhaps indirectly as a result of an unbalanced application of the resulting technology.

C. P. Snow identified the information explosion with the barrier of communication between the scientist and humanist; the gulf between the two cultures. In his erudite monograph Foskett (1) has examined the increasing lack of communication between the scientist and humanist from the point of view of the professional librarian: One faced with the task of trying to maintain information retrieval in a world whose boundaries, like those of the expanding universe, are lost forever to the observer's telescope. He comes to the conclusion that "Scientists tend to assume airs of arrogant superiority over non-scientists...control over material phenomena is possible to an extent undreamt of even fifty years ago, and rightly used, the discoveries of science could bring about that revolution in our material conditions foreseen by Wordsworth, who put the poet at the side of the man of science ..... There is no hope of such a creative partnership while scientists fail to carry out their duty of making these discoveries familiar". If the scientist does give this impression of himself it is a reflection of his education; a result of not seeing his fellow men and his environment in the very perspectives dictated by the world of science. This, in turn, is because we are educating technicians rather than scientists. We lose sight of the wood too easily for the technical trees.

Are not university courses terribly cluttered with unnecessary or even obsolete technical knowledge? Are we not attempting the impossible by trying to contain forever an exploding volume of knowledge? I suggest that the problems of documentation and information retrieval must play a much more vital role in scientific education. This would facilitate elimination from the syllabus of certain knowledge once documented and rapidly available through efficient information retrieval. It would give more scope for original thought; a chance to examine some of the fundamental problems of our time.

Let us adjust our perspectives. Geologists tell us that the earth is of the order of five billion years old. In order to grasp this time scale let us suppose that the earth was formed on the occasion of the birth of Jesus Christ, 2,000 years ago. Now on this scale William Caxton printed his first book just under three hours ago. The Wright brothers made their first powered flight ten minutes ago and 90% of all the world's scientists have been born since! The world population also doubled in the last ten minutes. We exploded the first atomic bomb less than two minutes ago. On this time scale the growth of scientific information and technology can indeed be seen as an explosion. It is an especially sobering thought if we try to look forward, just ten minutes!

Now let's turn our attention to space. Hoyle (2) considers it probable that there are one hundred thousand million stars with

planetary systems in our Milky Way galaxy alone. Hence "The probability of there being intelligent life 'out there' is overwhelmingly high". Hoyle has seriously suggested that with radiotelescopes little more sophisticated than those already in existence we should be able to establish a range of communication to embrace the nearest million stars. Somewhere in the million, Hoyle suspects, there are planets on which has evolved intelligence comparable or superior to our own. He has speculated that intelligent radio-communication may have been in progress for millions of years. If indeed we can tap such a cosmic reservoir of intelligence, get into the galactic telephone directory as Hoyle puts it, then our own information explosion becomes a mere bubble.

Is it easy for the scientist to conjure up feelings of super-iority or arrogance with this picture of his environment? Certainly not if this sort of cosmological appreciation were part of his educa-tion. In this way he could better approach the larger problems of humanity with essential humility. The humility due to the constant knowledge of our colossal relative ignorance.

What do we ask when trying to assess a candidate for appoint-ment or promotion? "How many papers has he published?" Perhaps we scan the titles or read a few summaries, lest we appoint a geneticist instead of a taxonomist! How often do we read even one of his papers from beginning to end? Not frequently. We haven't time. So the young scientist with an eye to attracting the attention of his peers gets out as many papers as he can.

We are all familiar with the appearance of substantially the same article in two or more journals. And there is another form of duplication: how many times do we read an almost identical descrip-tion of some well established experimental procedure such as this:- twenty grams of tissue were representatively sampled and accurately weighed into a Soxhlet extraction thimble and extracted for 24 hours with A. R. benzene. The extract was taken to dryness on a water bath and the non-volatile residue weighed etc. etc.

Before publishing we should first ask if we will contribute either to the knowledge which the student should embrace or to that to which the specialist should have access. If the answer be no then we should abstain even though it would give us our century. If yes, then how can we strip the publication of non-essentials? Is it to be a work of literature or a scientific communication? Surely the latter. One way of improving scientific communication would be to devise a kind of international shorthand. Some abstracting services have started this but we could go a good deal further so that the Soxhlet extraction paragraph might read something like: - Weight non-vol 24hr. C_6H_6 extr. 20 g tissue. Many consecutive operations such as those involving extraction, fractionation, detection and assay might well be indicated by a symbolic flow sheet. An International Conference to formulate such a shorthand based on English would be a most valuable contribution. Once terms and expressions were agreed upon, they could be published by the various learned societies in their Journals and their use insisted upon as indeed many abbrev-

iations already are. What I'm suggesting is that it's high time we regarded routine scientific publication for what it is: communication and documentation; not a work of literature.

Society is becoming increasingly dependent upon science and technology in a world of limited resources and dangerously unstable international relationships. This is clearly appreciated by politicians and administrators but the present tendency is merely to impose administrative or political philosophies on the world of science. The converse would be more to the point. That is, the philosophy of science, an absolute respect for the truth, might be profitably applied to the problems of government and administration and even, perhaps, to commercial advertising.

International instability has become a universal threat. These problems are a direct result of the impact of science on society. They require scientific analysis and control in a spirit of scientific humanism. Meanwhile, the best we can hope for is to keep open the communications between the nations. The machinery for this exists through the United Nations and its scientific or specialist agencies. The scientists of the world speak a common language and must subscribe to the same respect for universal truths. They have the best opportunities for international meetings and social and professional intercourse.

They must learn to contain their information explosion; to re-examine urgently the whole structure of scientific publication. Only then will they have the time to regain sight of the wood for the trees. The education of every scientist should provide for an objective scientific appreciation of his human and physical environment and the impact of his own technology. He will then be in a position to regulate better the production of "dangerous knowledge and disorganization" and to challenge its political abuse.

Unless every scientist emerges from the swamp of his own information he may indeed find himself continually on tap but never on top: an increasingly dangerous world will remain the politician's toy!

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AN APPROACH TO A PROBLEM IN POPULATION DYNAMICS*

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This is the text of a lecture to a group of graduate students in zoology and entomology. It describes the first stage of an investigation of the population dynamics of Malacosoma pluviale *(Dyar); what led to the problem; how the study was planned, and how it actually developed. Some examples show that previous experience may be used to advantage during the planning stage of an investigation, and that it also may help to exploit the first break-through that occurs. But another example shows that previous experience then may be a handicap, as it may keep one from seeing things as they really are. Thus, the second break-through in a new field is more likely to be accidental, no matter how deliberate it may seem in retrospect. In other words, research still progresses more erratically than our final reports suggest.*

This is not the kind of paper one expects to find nowadays in a scientific journal. It is not a straightforward account of methods, results, and conclusions. Instead, it is a discursive personal account of the beginning of one investigation, and its attendant difficulties and mistakes. It was originally a lecture given to graduate students and faculty of the Departments of Entomology and Zoology of the University of Alberta in 1961. I chose this approach because I thought students should hear at first-hand how our investigations really develop chronologically, and not always in the logical way in which we report them. I wanted to show what prompted the investigation in the first place, and how its first important turning-points were reached.

The lecture was to be published, but has been withheld until now because some of its points depended on data presented in an accompanying lecture, and this supporting material had to be developed differently for publication. Now that the data are in print (Wellington 1964, 1965) there is no longer any restriction on the content of the original address. The factual material is drawn from my investigation of the population dynamics of the western tent caterpillar, *Malacosoma pluviale* (Dyar).

Most research papers show investigators moving in such straight lines that one feels they often must have known their conclusions before they obtained their results! It is unfortunate that published reports so consistently give this impression. They do so, of course, because space limitations in journals permit authors to describe only the ideal routes to discovery. The truly erratic paths that lead there, or the first faint sign-posts that indicate the most likely route are almost never described. As the limitations imposed here are not so severe, I can tell you not only about my destination, but also something of my reasons for going and my ways of travelling there. There must be some sort of outline to which we can refer, however, so let us see how a straightforward description of the early work might be summarized....

In 1955, an outbreak of the western tent caterpillar was nearing its peak in the Saanich Peninsula of southeastern Vancouver Island. Because it offered an opportunity to study the effects of behavioral and climatic variations on the insect's population dynamics, I collected some eggs from the outbreak for experimental purposes, and also mapped its boundaries so that I could follow later changes in its extent.

In 1956, when the eggs hatched, I subjected the emerging larvae to a very simple activity test that exploited their response to light. This test revealed several types of larvae that differed in their ability to perform directed movements when they were separated from their fellows. Some were well-directed and active, others were disoriented and less active, and some were so sluggish that they scarcely moved. Controlled rearings showed that these differences were persistent, and that they also affected individual development and survival, because the various types of larvae differed in their ability to find and utilize food.

Artificial colonies composed of varying proportions of active and sluggish larvae were established, and their habits were compared with those of natural colonies in the field. These comparisons led to the identification of different types of natural colonies, and this discovery in turn enabled me to find areas where either active or sluggish colonies predominated. Once these areas were located, working hypotheses could be developed to account for their existence and predict the ultimate fate of the populations within them.

The first results suggested that behavioral differences may have a greater effect on an animal's population dynamics than theorists hitherto have supposed. But to establish this point it was necessary to subject the deductions arising from this thesis to repeated tests. Such testing has been the primary objective of the study since 1957 and, to date, accumulated observations tend to support the thesis in a most consistent way. For example, active individuals predominate in new infestations, but the sluggish component of the population increases as infestations age. Ultimately, most members of one generation are so sluggish that they cannot survive. Consequently, numbers within infestations so affected are drastically reduced.

Although very condensed and incomplete, this summary is sufficient to provide us with a framework for future reference (see also Wellington 1957, 1960). But why should anyone want to study the effects of individual differences in behavior or activity on a whole population? And if they must, why use the western tent caterpillar instead of some other animal? Furthermore, what led to the rather unusual method of separating the different types of larvae at the beginning of the investigation? And finally, though the summary seems tidy enough, was the progress of the work really so direct? Or was it sometimes saved accidentally from ineffectual circling? In the remainder of this lecture, I will try to answer these questions.

To answer the first three I must go back several years before 1955. Those of you who read population literature know only too well the continuing debates among the theorists. For those who are less familiar with this literature, I can summarize its central theme in the following way. Many animals are alternately scarce and plentiful. Their numbers

increase tremendously for a few generations, then decrease again. A major problem for economic zoologists is to find out what prevents their indefinite increase; and bad weather, exhaustion of food supplies, or overwhelming attacks by enemies are often given as reasons why populations decrease. The situation is not so simple, however, because the numbers of animals may continue to decline while the weather is favorable, and while food is abundant and enemies are scarce (Chitty 1960).

Although population theorists often disagree, such conflict would be welcome if it included suggestions for *experiments* designed to *disprove* hypotheses. More often than not, however, it involves only comparisons of all-embracing theories. At least this is how it seems to field ecologists, who also find a disturbing gap between what the major theories say should happen in the field, and what actually happens there. Many investigators therefore have been dissatisfied with population theory for a long time.

Before 1952, I was too preoccupied with studies of the effects of weather on the behavior of insects to be concerned with the theory and practice of population ecology. One cannot study the effects of weather on insects for long, however, without being drawn into some of the population controversies. But when I finally began to consider the various arguments, I found I was less concerned with some of their more evident misinterpretations of weather processes than I was with the way in which they neglected the behavior of animals.

My own experience made me notice an operational weakness in most studies of population dynamics. In many of these studies there was a tendency to concentrate on the developmental and reproductive processes of the animals, and on measurements of their mortality or survival, to the virtual exclusion of their behavior and activity. But this approach overlooked the stubborn fact that an animal that does not behave properly, or that does not maintain a certain level of activity at critical periods in its life, simply does not survive, let alone develop and reproduce.

The more I thought along these lines, the more I felt that the right kind of observation would show that widespread neglect of the influence of individual behavior on survival was actually obstructing the development of population theory. And this feeling was not just a product of the scientific chauvinism that might be expected from my studies of behavior; it arose from the observation that some of the major theories could not really be falsified in their existing form (c.f. Platt, 1964). This was my main reason for wanting to study the effects of the activity and behavior of individuals on the fate of a whole population. But I had to find an insect that would be suitable for such a study.

I had one hint from previous work that *Malacosoma* spp. might be suitable. In 1948, C.R. Sullivan and I had studied the light reactions of three species of *Malacosoma* that were prevalent near Sault Ste. Marie, Ontario. We were interested in the changes in response that might take place at high temperatures. And we had been following the usual procedure; scattering larvae at random on the platform of a choice chamber that had illuminated and darkened sides. The insects were expected to

take up positions dictated by their initial response to light at room temperature, then move to different locations if their response changed when the temperature was raised.

We had done virtually the same thing with other kinds of insects many times before. But when we used newly-emerged first-instar larvae of *Malacosoma* only a few acted in the expected way. The majority never moved after they were dropped on the platform. Consequently, we could not continue the experiment, because we could not tell how they reacted to light.

To solve this problem, **we put the** larvae back on their egg mass, so that they would be in a more natural situation. In effect, we made the egg mass the dark-light alternative, with its top illuminated and its bottom shaded. When all the larvae were allowed to remain together on their eggs in this way, they moved about very easily. And since this solved the technical problem, we proceeded with the investigation (Sullivan and Wellington 1953).

I wondered afterwards, however, why most members of these young colonies could perform directed movements while they were touching one another, but not while they were isolated. And if most of them were so dependent, why were a few so independent that they could perform directed movements while they were alone? I had to file this puzzle for future reference, however, because we had used all the available larvae. And eventually, of course, I stopped thinking about it.

But I remembered it again in 1952, when I began to think about the possible effects of individual behavior on a population. Here, apparently, was a group of insects that varied in activity and behavior as soon as they hatched. Besides, all the members of the genus also experienced great and comparatively regular changes in numbers. And some species made conspicuous tents, so that they could still be found without much difficulty when they were scarce. *Malacosoma* spp. thus had much to offer as experimental animals.

It was no help to realize this in 1952, however, because the tent-forming species were too scarce to provide enough material for testing. But when I saw the outbreak of *M. pluviale* on the Saanich Peninsula in 1955, I was again reminded of my earlier intentions, and pleased to see a good supply of one of the species that had provided the germ of the idea. And that is how *M. pluviale* became the experimental animal in the study.

It is worth noting that at this stage I had very little foundation on which to build a work plan. I knew nothing of the apparent difference in activity that I have just described, except that it existed. I did not know whether it was simply an intrinsic part of each individual's make-up, varying from time to time as the animal passed through different physiological states, or whether it was a real and persistent difference among individual *Malacosoma* larvae, stable enough to be exploited in the type of study I had in mind. Since it would not take long to find out which kind of variability was involved, however, I decided to plan the forthcoming investigation on the assumption that the difference would prove to be persistent.

The decision to plan the investigation in this way did not depend entirely on an act of faith. I had recently observed peculiarities in the

behavior of some arctiid larvae which suggested that such individual differences might in fact be stable. Also, as I came to realize later, my various lines of thought had been channeled during a brief conversation with Dennis Chitty just before I saw the tent caterpillars on the Saanich Peninsula. Thus my ideas concerning individual behavior were resting comfortably within a larger framework. And larger frameworks are always reassuring, even when one is scarcely aware of them.

During our conversation, Chitty and I discovered we were both dissatisfied with current population theories, and disturbed by the tendency of ecologists to treat the populations with which they worked as though they were monolithic structures, instead of collections of individuals. But Chitty also was circling an idea he has since stated more explicitly; namely, that the composition of a population might change with changing density, and that this qualitative change might have important effects on subsequent densities (Chitty 1960). Looking back, I do not believe I had carried my ideas about the effects of individual behavioral differences on populations quite so far (although my ready response to Chitty's well-nigh subliminal prompting showed me later that I had obviously been ready to do so). A few months afterwards, however, all that was clear to me at the beginning of my own study was that I not only had to determine how any variations in behavior might affect the survival of individuals within a population; I also had to consider these individual differences in terms of the changes in population quality with which they might be associated. Still later, when I had some results to interpret, I suddenly realized that my final plan of attack had been decided, virtually at the last minute, by that conversation with Chitty: a conversation, incidentally, that I had "forgotten" in the enthusiasm engendered by finding the *Malacosoma* outbreak and planning my investigation.

The first step in that investigation was to ensure that the differences observed in 1948 were truly persistent between individuals, not just internal changes within any individual at different times of the day or between successive days. If the former situation obtained, many things followed directly. Otherwise, I scarcely had a problem of the sort I had imagined. To establish the facts, repeated tests of identified individuals were required. And I needed a very simple and rapid screening method that would allow me to handle large quantities of material; e.g., perhaps more than 15,000 larvae per generation. It seemed best to exploit the difference in activity noted during 1948, as it appeared to be present as soon as eclosion took place. This, then, was one reason for using the laboratory test employed at the beginning of the investigation. But there was another reason that requires further explanation.

Some aspects of reality are unusual enough to seem unacceptable or even unbelievable when we first encounter them. In these days of team research and elaborate equipment, we tend to forget that explication of these unusual and often complex aspects of reality does not always require a complicated attack. In fact, some of our more mechanized attacks only obscure reality, or the approaches to it. And obscuring the path to an incredible result does not often encourage others to verify or disprove it.

A good example of what I mean may be found in Karl von Frisch's work on dancing bees (1950). Some of those early results and conclusions were quite unbelievable, but the experiments had a truly beautiful simplicity. Without such simplicity, other scientists might still be questioning von Frisch's conclusions. Because of it, they have been busily extending his results; though, unfortunately, not always with such elegant methods. Present-day biologists have much to learn from Professor von Frisch's approach to problems, therefore, and can profit from it in whatever field they intend to explore.

I was prompted by this line of thought to devise a very simple test for my own purposes. As each egg mass hatched at room temperature, I took its newly-emerged larvae and distributed them in a long line parallel to a fluorescent lamp, separating the individuals so that they had to move more than their own body length before they could touch any of their fellows. The reasoning was that any individual capable of independent, directed movement should proceed directly toward the light, whereas the others should stay where they were, or not move very far in any direction. This should separate any colony into at least two components. And the stability of each of these components then could be assessed by further testing.

The test worked very well. It was in fact my first breakthrough, because without such an easy, rapid, and definite means of identification of persistent differences among individuals, there would have been little time to do anything else. Because of the test and its results, however, the first part of the study opened automatically into a series of sub-projects that virtually had to develop along certain lines, often with results that were quite predictable, because they were the logical outcome of the existence of the behavioral differences.

Consider the results of the rearing experiments, for example. Larvae that differ in their ability to perform directed movements must behave in certain predictable ways when they are gathered into groups and placed near food. Very sluggish larvae should be incapable of fending for themselves, no matter how many are grouped together. And this proved true. Very sluggish larvae had to be placed on their food because they were incapable of locating it when there was no active individual to guide them, even when the food was only a few mm. away. Without proper care, therefore, they starved. And proper care included frequent inspections to ensure that they had not fallen from the food, because they could not return to it unaided.

More active, but still disoriented larvae proved relatively easy to handle, as long as they were kept in sufficiently large groups. Then they spun sufficient silk to be protected from desiccation, and they eventually found food by a sort of group "amoeboid" flow. Thus they fed and developed, though with some delay.

In contrast, the independent larvae were more difficult to handle under artificial conditions. They were too independent in the rearing jars; a predictable result of their ability to orient and

travel while isolated. Although each could find food very quickly, individuals tended to remain scattered for hours instead of clustering together occasionally. Therefore they had few opportunities to form the common mat of silk that would protect them from desiccation, so that they often died when only small numbers were kept together in the jars. Increasing the number of larvae per jar, however, solved this problem.

As development proceeded, it was clear that the most active larvae fed more and developed most quickly, whereas the most sluggish, if they lived at all, fed least and grew most slowly. There was no evidence within the generation that disease or any malfunction not attributable to the basic differences was at the root of such variation. There was plenty of evidence, however, that eggs laid by some females yielded colonies that had a high proportion of *sluggish* larvae, whereas eggs from other females yielded colonies that had a much greater proportion of *active* larvae.

Many other differences in behavior and activity were revealed during these studies, which opened endless avenues for further physiological research. But I must confine my remarks here to the development of the population studies. The foregoing descriptions were necessary to emphasize that there were some very marked differences in development and survival associated with the differences in activity and behavior, even though the latter were first revealed as an apparently trivial response.

As the rearing experiments with pure groups progressed satisfactorily, I began to make up artificial colonies differing in the proportions of the types of individuals they contained. These were studied in the laboratory and in the field to determine what differences in growth or habits they might have. Those which contained numerous well-directed larvae were active. They formed several tents in rapid succession, spacing them widely over the available foliage, and vacating each in turn before they exhausted the food nearby.

In contrast, colonies that contained a high proportion of sluggish individuals were very inactive. Such a colony seldom made more than one tent, and the larvae spent much time clustered on it, because there were not enough active individuals present to disturb and scatter the other larvae resting in the cluster. The larvae enlarged the tent and occasionally fed out from it for short distances, but even when they had exhausted nearby food they seldom moved on to spin another tent, though ample food was available only a short distance away. Consequently, the members of truly sluggish colonies usually starved. If they were saved from this fate by unusually abundant food right at hand, they were still prey to disease. (They were more exposed to infection than members of active colonies, because they often touched the remains of diseased larvae during their prolonged clustering periods.) Very sluggish colonies, therefore, soon were lost to the population by one or other of these means.

When I finally obtained adults from the different types of larvae, I found that activity differences were still recognizable, and that their classification could depend once more on a very simple

test. Active adults left in the jars in which they emerged literally battered themselves to pieces in one or two days. From this extreme there ranged a graded scale of decreasing damage to the other extreme: the perfect appearance of sluggish adults that remained unmarked until they died. They never moved after their wings expanded.

All the findings described above came from straightforward exploitation of the logical consequences of the original differences observed among emerging larvae. They were necessary steps in the study, but most of them could not immediately add to its further development. As an isolated group of facts they offered no direct entry into the next stage: the study of the natural population. In fact, while all these sub-projects were in progress, I had been trying to find a way to distinguish the different types of natural colonies in the field without having to classify every larva within them. Without a simple and rapid method of classifying the natural colonies, I could not progress with the field studies.

The artificial colonies finally provided the solution to this survey problem. For not only did the active colonies among them make more tents than the sluggish colonies; they also made tents of a different shape. The "active" tent was longer and thinner -- in most instances very obviously club-shaped -- whereas tents made by less active colonies were shorter and squatter; in extreme instances, definitely pyramidal.

Here I had the potentially perfect sorting method to bring order out of the apparent chaos of the peak population of 1956, provided that natural colonies behaved as the artificial ones had. If they did, I could close the gap between laboratory and field studies by using differences in tent shape as a simple but reliable survey tool to classify every colony I examined. With it, I should be able to see whether there were areas where one type of colony predominated. In addition, I should be able to accumulate statistics on differences in the sizes of feeding areas, larval numbers, etc., among colonies. I also should be able to identify colonies that had changed their characteristics during development after losing one or other of their constituent groups, because these changes should be revealed by differences between their previous and current tests.

With so many potential benefits due, I was almost afraid to examine natural colonies again in case the difference did not exist among them. It was there, of course, as it had been all along. I had not seen it before, however, even though I had been happily finding and counting colonies by watching for their tents! I did not see it because I had been caught in the snare that lies in every research path: inability to get outside one's previous conceptual framework. Because every entomologist *knew* that tent caterpillars occupied box-like or pyramidal tents, I had paid no attention to tent shapes in my earlier surveys. Consequently, I saw them properly only after I had a strong incentive to look.

This second break-through of the investigation was a happy accident, therefore, and not the product of deliberate planning that

the first had been. If it had not occurred, however, not much else would have happened during that first season of study, and I would have begun the next with a serious handicap. Consequently, I have emphasized it and the preceding mistake. In fact, this whole sequence of events is a good example of the greatest difficulty that confronts us whenever we engage in frontier research. At the border of the unknown, one *must* consciously strive to escape from the mesh of former frames of reference, and to remain outside the generally accepted range of opinion concerning one's problem, for a very good reason: the problem is rarely what accepted opinion says it is! But the difficulty is that one tries so hard to keep one's thinking free on larger issues that one overlooks the danger of continuing to think about apparently smaller issues in terms of older concepts. This lapse is always dangerous, and sometimes disastrous, because there is *no* small issue at a frontier. And how can one *observe* what does not yet exist as a conceptual possibility (Hanson 1958)?

A new survey soon showed that club-shaped tents predominated in areas that were unoccupied by the expanding population before 1956. In fact, if the new infestations of 1956 were sufficiently far from previous infestations, only club-shaped tents occurred. On the other hand, a larger proportion of pyramidal tents occurred wherever the population had been in residence for several generations. In such areas, some trees contained only pyramidal tents, although there were always some club-shaped tents in any locality.

This information led directly to a testable hypothesis concerning the fate of any local population after its first introduction into an area. It seemed reasonable to suppose that active adults would, in general, produce active colonies, whereas less active adults would produce colonies that were decreasingly active, down to a level where some would be very sluggish. Also, it was already known that these various types of adults differed in their ability to fly. Further observation of their movements made it clear that only the most active could fly far enough to enter remote, previously uninfested areas. Therefore, in a new, remote locality, only active colonies should be produced by these first invaders.

Provided that survival within these colonies was adequate, however, adults that displayed different amounts of activity would be produced from them (since even active colonies contain some inactive or sluggish individuals). Of these, only the active adults would be able to fly away before they oviposited; the less active would have to oviposit closer to their birthplace. The next generation in that locality, therefore, should contain some colonies less active than any of the parent generation. And in subsequent generations, an increasing proportion of sluggish colonies should appear in the locality if emigration of active adults exceeded their immigration. This is what the local differences observed in 1956 suggested, and it remained to be seen what actually happened after 1956.

As working hypotheses go, this first model turned out quite well; i.e., its major statements could not be disproved. Certain aspects of the general population trend and of the local environment

affected the situation in any locality. But within these limitations, only minor amendments to the hypothesis were required. When newly infested areas were sufficiently remote, the first generation in fact consisted entirely of active colonies. In contrast, new infestations established closer to older ones contained some less active colonies in the places nearest the older foci -- a fact, incidentally, that helped to establish maximal flight distances for less active females. In the next generation in an isolated area, however, some sluggish colonies appeared, and their proportion rose during subsequent years until the population included many colonies too sluggish to survive. Similar changes, though further advanced, could be recognized in older infestations. The end result was always the same: a sudden reduction in numbers, because most of the colonies had died.

In that last paragraph I hurried through the findings of several years, after using considerably more space to outline the sequence of events that led up to them. But this is as it should be, if I am to fulfil the intention outlined in my introductory remarks. All the foregoing results have been published, along with many others I have not mentioned here (Wellington 1957, 1960; 1964, 1965). But until now, I have not described how I reached them. And it is reasonably correct to say of this, as of all scientific work, that most of the original thinking had been done by the time the first experiments were completed. After 1956, the speculation and reasoning that had led to the first tentative proposals were buried by the pedestrian process of testing them.

Finally, I should point out something not emphasized earlier, though it is implicit in much of the foregoing description. Although this was, and is, a field study of a population, the laboratory has had a strong influence on its inception, direction, and findings. My original dissatisfaction with population theory and practice stemmed partly from the fact that laboratory studies of insect behavior made me sceptical of some of the ideas and conclusions of population ecologists. Many of the clues on how to approach the problem I wanted to investigate came from laboratory observations, as did the evidence for the initial differences. Similarly, the different tent-shapes were detected only by studying colonies with controlled compositions; a method that is still more common in laboratory studies than it is in the field.

And this brings me to the point I wish to make. I believe that laboratory studies by themselves often degenerate into the pursuit of trivia. But I also believe that field studies that lack the benefit of the special discipline that comes from laboratory training and planning are unlikely to advance much beyond the speculations with which they begin. In other words, the theory and practice of population ecology should not be exempt from the general rule that hypotheses are better disciplined by experiment than by faith and reason (Chitty 1957). Consequently, when we cannot combine laboratory and field studies during population research, we should at least take the discipline of the laboratory with us when we go to the field.

A balanced program of laboratory and field investigations in

fact has some very practical attractions. In the studies described here, I was able to do much more during the 1956 season (a matter of some two months) by keeping the laboratory stocks and tests slightly ahead of the equivalent stages in the field. Thus I was able to make any number of mistakes during the first round of experiments and observations, and still have time to correct them by using fresh material as the field population entered each required stage. This enabled me to exploit the two break-throughs of that first season with minimal delay.

ACKNOWLEDGMENTS

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POPULATION STUDIES ON EDMONTON MOSQUITOES*

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The seasonal fluctuations of each instar larvae and pupae of Culiseta inornata (Williston) in a particular pool near the University of Alberta were investigated and an attempt to estimate the mortality of the aquatic stages was made. The data for the collections of adults and larvae of 26 species of mosquitoes found around Edmonton indicate that the black-legged mosquitoes of subgenus Ochlerotatus, *genus* Aedes *are earlier-appearing species than others. The distribution pattern of mosquito larvae was firstly demonstrated to follow a negative binomial distribution with a common value of constant k for various density levels. Based on this distribution pattern, a sequential sampling technique was applied to classify a mosquito population into one of three predefined density levels. This was considered useful in deciding whether or not control is necessary, and in evaluating whether or not control has been successful over a wide area in a relatively short time.*

GENERAL INTRODUCTION

The City of Edmonton has been engaged in the control of mosquitoes and has reduced the mosquito population greatly in the city (see Klassen and Hocking, 1963 and 1964). However, there are still some problems to be solved. They include precisely when and how the insecticidal applications should be made for the effective and economical control of mosquitoes, how far the larvicide should be applied beyond the city limits, and so on. For the settlement of them, extensive fundamental studies are required. This report deals with the studies conducted in 1964 to approach the problems from an ecological point of view.

BICNOMICS OF EDMONTON MOSQUITOES

Mosquito Surveys and Identification

Three types of mosquito surveys were made in 1964. Firstly larval (and pupal) surveys were made at pools in various environments around Edmonton, mostly westward, from April to July. The number of dips at each pool was not recorded, except for a few pools for determining the distribution pattern of larvae per dip, which will be mentioned later. However, care was taken in catching mosquitoes so as to represent the mosquito fauna there; only a few dips were made at pools with high mosquito density and many dips, sometimes more than 50. at pools with low density.

Secondly collections were made of adult mosquitoes, which came to feed on me, around a particular pool near the University of Alberta at approximately one week intervals.

Thirdly larval surveys were made at the pool mentioned above. The pool harbored almost exclusively *Culiseta inornata* (Williston) and the seasonal changes of immature stages were studied.

The larvae collected were reared in the laboratory to the fourth instar or to adults, and identified. Some specimens were separately reared to obtain the adults with associated larval skins to facilitate determining the species.

The identification of larvae followed Carpenter and La Casse (1955) and Rempel (1950). Adults were identified mostly after Carpenter and LaCasse (1955) and Rempel (1953). However, it was often difficult to separate them to species, especially rubbed specimens of black-legged female *Aedes*. In such cases, and even for good specimens, the post-coxal scale patch (between the anterior coxa and the sternopleuron), mesepimeral scale patch, scales of probasisternum, and tarsal claws were useful characters (Beckel, 1954; Vockeroth, 1954).

Notes on Some Species

Aedes communis (DeGeer) and *Aedes intrudens* Dyar

A. communis and *A. intrudens* are black-legged species lacking the post-coxal scale patch. The adult female of *A. communis* is usually separable by the contrasting stripes on the scutum from *A. intrudens* with a uniformly colored scutum. However, in some specimens of *A. intrudens* the scutum shows indications of paired median brown stripes, and those specimens, particularly when the scales on the scutum are not complete, are sometimes hard to distinguish from *A. communis*.

After examinations of 38 females of *A. intrudens* and 32 of *A. communis*, some of which were associated with their larval skins, it was found that, as described by Carpenter and LaCasse (1955), mesepimeral scales reach near lower margin in *A. communis*, but in *A. intrudens* the lower third or fourth is bare. This seems to be a most useful character to separate them. Other characters, which might be used, are the number of lower mesepimeral bristles and the color of the base of the costa. The lower mesepimeral bristles vary in number in both species, but, in the present specimens *A. intrudens* has a smaller number of bristles, ranging from 0 to 3, than *A. communis*, which has 2 to 7 bristles. White scales at base of the costa are absent, or if present very few in number, in *A. intrudens*; they are present in *A. communis*.

Aedes hexodontus Dyar and *Aedes punctor* (Kirby)

The adults of these two species are very similar to each other, however the larvae are distinct. According to Beckel (1954) the probasisternum has white scales and an extensive patch of white

scales is seen at the base of the costa in *A. hexodontus* taken in the field at Churchill, Manitoba; on the other hand in *A. punctor* taken there scales on the probasisternum are reduced to a few and there are no white scales at the base of the costa or rarely one or two. These characters were found useful to separate specimens of these species taken near Edmonton also, by examination of females associated with their larval skins.

Knight (1951) recognized two varieties in each species: "type *hexodontus*" and "tundra" variety in *A. hexodontus* and "type *punctor*" and "tundra" variety in *A. punctor*. The scutum of females has a broad median dark stripe which may be narrowly divided in "type *hexodontus*" and "type *punctor*", on the other hand in "tundra" variety of both species the median dark band is absent or not well defined.

Of the females of Edmonton *hexodontus* collected or reared from larvae, 9 are "tundra" variety and one is "type *hexodontus*" variety. The latter was collected as an adult on June 7, 1964. In addition to these, I have another female specimen of "type *hexodontus*" variety, which was reared from a larva taken near Jasper, Alberta, on May 16, 1964. The associated larval skin shows that head hairs 5 and 6 are both double, which agrees with the description given by Knight (1951) for "type *hexodontus*" variety.

As for *A. punctor*, the many larvae and 18 females, which were collected as adults or reared from larvae, are considered all "type *punctor*" variety.

Aedes niphadopsis Dyar and Knab

A larva of this species was taken from a collection of small scattered pools in a pasture near a creek, about 20 miles west of Edmonton, on June 7, 1964, and reared to a female adult. This record is new to Canada (Pucat, 1964).

Aedes pullatus (Coquillett)

This is a species that lacks the post-coxal scale patch, and bears a distinct hypostigial scale patch. The distribution in Alberta seems to be limited mostly to mountainous regions. I collected many larvae from snow-melting pools in Jasper National Park on June 21, 1964, but no specimens were encountered around Edmonton.

Seasonal Fluctuation and Mortality of Immature Stages of *Culiseta inornata* (Williston)

Observations were made on the changes in abundance of each instar larvae and pupae of *Culiseta inornata* (Williston) throughout a season at approximately one week interval in 1964 at a pool, ca. 10 x 3 m, near the University of Alberta. The pool is situated on the south bank of the North Saskatchewan river, and receives little sunlight because of tall vegetation such as poplars around it. For this reason, ice remained at the bottom of the pool as late as May 8, and the water temperature was relatively low throughout the summer; the maximum water temperature was only 18.3 C, on August 17.

On each day, larvae and pupae were sampled with a dipper usually ten times, but when necessary, 20 or 50 times, and the

numbers of each instar larvae and pupae were recorded. The population of mosquitoes in the pool consisted of only *C. inornata*, as far as the fourth instar larvae were examined. However, from some egg rafts collected at the pool on July 6, there emerged some adults of *Culiseta alaskaensis* (Ludlow) in addition to *C. inornata*; this indicated that a small number of egg rafts, probably one, of the former species was mixed in the collection of the egg rafts. Therefore, some *C. alaskaensis* may have bred also in the pool, even so, the number seems to have been negligibly small.

Egg rafts were first encountered on May 25, and oviposition continued until August 10. The number of egg rafts per dip and the observation for the rafts on the water surface of the pool show that the peak of oviposition activity of *C. inornata* was in the first half of June.

The seasonal distribution for each instar larvae and pupae is shown in Fig. 1. The first individuals of larvae in the first, the second, the third, and the fourth instar, and pupae were encountered on May 25, June 2, June 16, and June 22, respectively. The peak in numbers of first instar larvae was June 8, and with the progress of the development the time of each peak became successively later; the peak for pupae was on July 6. The period between the peaks of first instar larvae and of pupae is about one month. This seems to be the time required for *C. inornata* to develop from the first instar larva to the pupa; the mean water temperature was 11 C during the period.

The emergence of adults is thought to have occurred most actively shortly after the peak of pupae, that is in the middle of July. This time of peak emergence was ascertained by the fact that many pupal skins were observed on the water surface on July 14 and 23.

It has been reported that the duration of the larval stage of mosquitoes such as *Anopheles quadrimaculatus* Say and *Aedes aegypti* (L.) is affected by temperature, nature and amount of food, and density of a population (e. g. see Horsfall, 1955). Therefore, the above period of one month at mean water temperature of 11 C will be changed to some extent according to the conditions in a pool, even when the temperature is the same. Also, the remarkable difference in water temperatures within a pool (Haufe, 1957) may influence the data. However, the difference does not seem to be great, as most larvae inhabit similar environments.

The area surrounded by the abscissa and the curve for each instar larvae and pupae in Fig. 1 is dependent on the relative abundance and also on the duration of each instar. In the laboratory at ca. 23 C, an egg raft of *C. inornata* was reared to adults, and mean periods for each instar larvae and pupae were obtained. If it is supposed that these mean periods are kept unchanged also in the present field data, we can get the relative abundance by dividing the calculated area from Fig. 1 by the mean period. The results are given in Table 1. It is recognized from the table that the reduction in the relative abundance is remarkable between the first and the second instar larvae, and between the third and the fourth instar larvae. The survival rate
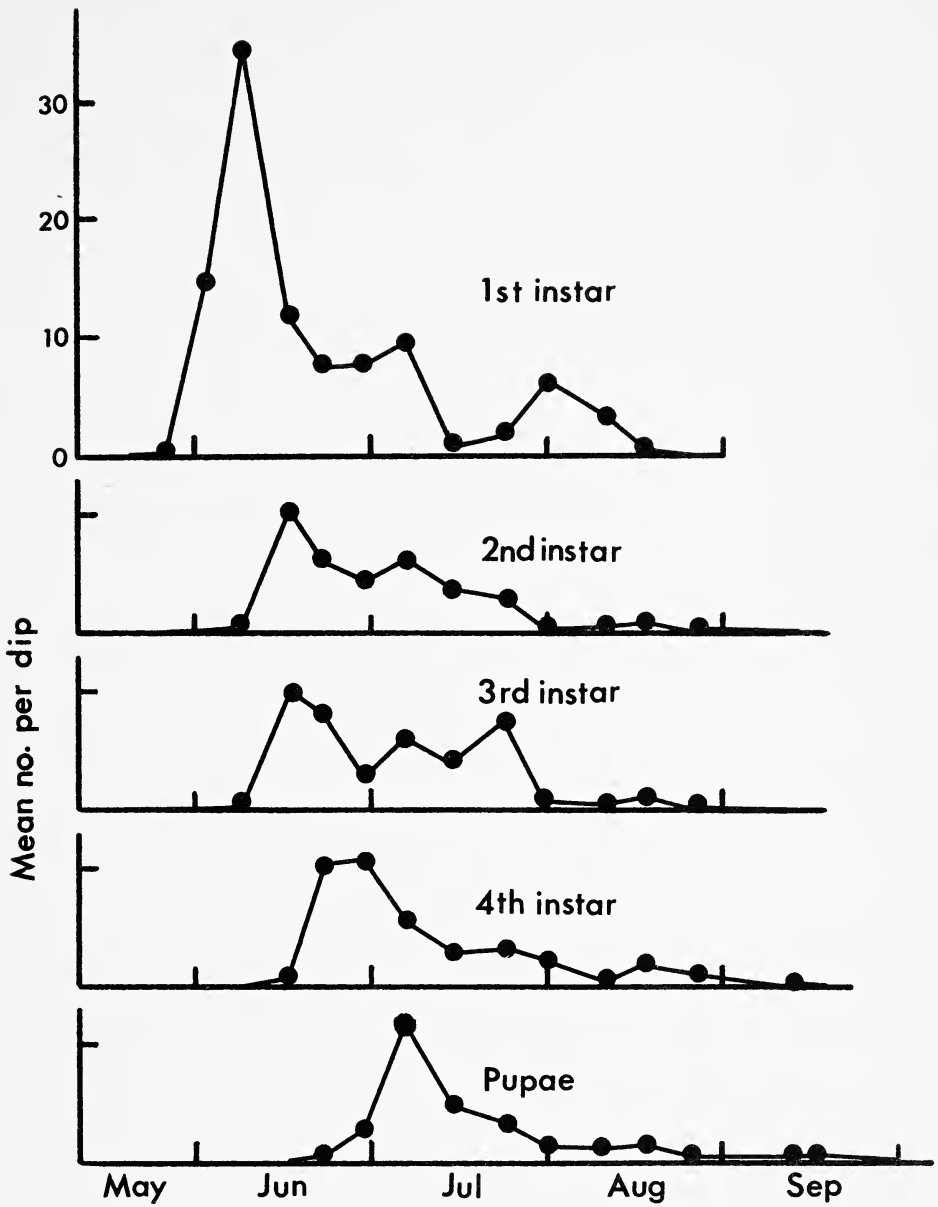
Fig. 1. Mean number of each instar larvae and pupae of *Culiseta inornata* per dip.

from the first instar larvae to the pupae is estimated at 63/248 x
100 = 25%. Thus we get a mortality of 75% for the aquatic stages of
C. inornata, or slightly higher, as the mortality in the earlier half of
the pupae is not included in the above calculation.

TABLE 1 - Relative abundance of each instar larvae and pupae of
         C. inornata in the field.

		Larvae				Pupae
		1st	2nd	3rd	4th	
Area (days x no. of individuals) in Fig. 1	(A)	718	275	311	292	220
Mean period (days) in the laboratory	(B)	2.9	2.0	2.3	4.2	3.5
Relative abundance in the field	(A/B)	248	138	135	70	63

The reliability of the above calculation depends on how
effectively the material was sampled from the pool and how close
the relative mean duration for each instar larvae and pupae obtained
in the laboratory is to that in the field. As will be mentioned later,
the number of larvae plus pupae of mosquitoes per dip follows a
negative binomial distribution having a larger variance than a random
distribution. This means that a larger number of dips is required
to estimate the population effectively, and the number of dips may
be too small in the present field data. As mentioned earlier, the
mean duration of larval stage is affected by temperature, food, and
population density, but perhaps little affected in pupae by the last
two factors. Therefore, it is rather difficult to compare the values
in the laboratory with those in the field. Another difficulty is that
the temperature in the field changes daily and seasonally. Never-
theless, the above method of estimating the mortality is of value as
a first approach to this important subject.

In any case, it seems that the mortality in the aquatic stages
of *C. inornata* is fairly high in the field. The factors responsible for
this are not known. However, physiological disorder or a sort of
disease is supposed, as some dead larvae were found and all attempts
to find predators in the pool failed.

Seasonal Occurrence of Edmonton Mosquitoes

Table 2 gives the number of larvae (and pupae) collected
around Edmonton and the number of collections in which each species
was found. Mosquitoes were encountered at 30 pools out of more
than 60 examined. Since the number of dips varies from pool to

pool, the number of larvae shown in the table does not represent exactly the relative abundance of each species. However, the main features of seasonal appearance are clearly seen.

TABLE 2 - The total number of larvae and pupae collected around Edmonton, and the number of collections (within parentheses) in which each species was found.

Species	April early	April late	May early	May late	June early	June late	July early	July late	Total
Anopheles									
earlei				58(2)		4(2)	8(1)		70(5)
Culex									
tarsalis								1(1)	1(1)
territans						1(1)	5(1)	3(2)	9(4)
Culiseta									
alaskaensis								1(1)	1(1)
inornata					20(1)	436(2)	7(2)	158(2)	621(7)
morsitans					2(1)				2(1)
Aedes									
campestris		37(1)							37(1)
canadensis		1(1)							1(1)
cataphylla	19(3)		3(1)						22(4)
cinereus		1(1)	3(2)	4(1)	2(1)				10(5)
communis	10(3)	34(1)	16(2)						60(6)
dorsalis				1(1)	1(1)	2(1)			4(3)
excrucians	5(2)	6(1)	6(1)	13(1)	2(1)				32(6)
fitchii	31(2)	65(2)	3(1)	47(1)	2(1)				148(7)
flavescens				3(1)	1(1)				4(2)
hexodontus	3(1)	3(2)	1(1)						7(4)
implicatus	23(3)	24(3)	20(1)	1(1)	3(1)				71(5)
increpitus	1(1)	33(2)		33(1)					67(4)
intrudens	40(2)				2(1)				42(3)
niphadopsis					1(1)				1(1)
pionips			1(1)						1(1)
punctor	42(3)	2(1)	12(1)		1(1)				57(6)
riparius	29(2)	27(1)	3(2)						59(5)
spencerii	6(2)		10(1)						16(3)
vexans			2(1)			126(4)			128(5)
Total	209(8)	233(5)	80(4)	160(2)	37(1)	569(6)	20(2)	163(2)	1471(30)

The results of the collections of female mosquitoes, which came to feed on me, around a pool on the south bank of the North Saskatchewan river near the University of Alberta are given in Table 3. This table also indicates an aspect of seasonal fluctuations of mosquitoes.

From these tables and some other data, seasonal occurrence of mosquitoes in 1964 is given below.

Anopheles

Anopheles earlei Vargas hibernates as an adult female. Many larvae were found from late May to early July (Table 2), and one female was collected at the campus of the University of Alberta on May 26. Most of 58 larvae collected in late May shown in Table 2 were in the second instar and a few were in the first and a few in the third. Thus it seems that hibernated females appear and oviposit their eggs from May, and the emergence of adults occurs from June. Oviposition continued at least until the beginning of July, as two first instar larvae were encountered in early July.

Culex

The species of *Culex* found were *C. tarsalis* Coquillett and *C. territans* Walker. Both hibernate as adult females.

Although only one larva of *C. tarsalis* was collected, the hibernated females are considered to oviposit late in the season, as it is reported that in irrigated areas of Alberta the larvae are found abundantly in July, August, and September (Shemanchuk, 1959), and in Saskatchewan the first larvae do not appear until early July (Rempel, 1953).

The larvae of *C. territans* were collected in late June to late July (Table 2), and this seems to be also a late-appearing species .

Culiseta

Three species were encountered around Edmonton, namely *C. alaskaensis* (Ludlow), *C. inornata* (Williston), and *C. morsitans* (Theobald). They all hibernate as adult females.

The first egg raft of *C. alaskaensis* was found on July 6, as mentioned earlier, and one larva was collected in late July (Table 2). According to Jenkins (1948), overwintered females were common from late April to mid-June and all instars of larvae were found from May 11 to July 10 in Alaska. Therefore, the larvae may appear earlier than July also around Edmonton.

Table 2 indicates that the larvae of *C. inornata* were collected from early June, and this agrees with the data mentioned earlier. The peak of oviposition was found to be in early June and the peak emergence occurred in mid-July. The feeding activity seems to be limited mainly to the period from late May to early July, as judged from the number of females attracted to man (Table 3), and this is justified by the time of the peak of oviposition. Those females are considered overwintered ones. However, a small number of females oviposited as late as August 10 as mentioned earlier. It is

not known whether such oviposition was derived from overwintered fe-
males or from newly emerged ones.

TABLE 3 – The number of female mosquitoes collected around a pool on
the south bank of the North Saskatchewan River near the
University of Alberta, Edmonton.

Species	May		June				July				Aug.		Sept.		Total
	19	25	2	8	16	22	6	13	23	30	10	25	11	16	
Culiseta inornata		1	1	1	1	1	1								6
Aedes															
cataphylla	1														1
cinereus						1		1			1	3			6
communis		1		1	2	2									6
excrucians						1									1
fitchii						12	2	7	1				1		23
hexodontus		1		1		1									3
implicatus	4	3		1		25	1								34
increpitus						3	1			2	1				7
intrudens			1												1
punctor				1				2				1			4
riparius						1		2	1		1				5
stimulans														1	1
vexans								3			3				6
Total	5	6	2	5	3	47	5	15	2	2	6	4	1	1	104

*
One hour collection was made in the afternoon each day, excepting two
hour collection on June 22.

Two larvae of *C. morsitans* were obtained in early June. Rempel
(1953) reported the adults in July. This species perhaps spends a similar
life cycle to *C. alaskaensis* and *C. inornata* in Alberta.

Aedes

All *Aedes* species recorded here hibernate as the egg stage.

Black-legged species belonging to the subgenus *Ochlerotatus* are generally earlier-appearing species than other mosquitoes. The dates of the collections of the larvae and adults in those black-legged species (from Tables 2 and 3), together with the records of the larvae and adults in Saskatchewan by Rempel (1953) and the dates of emergence near Edmonton by Klassen and Hocking (1964) are shown in Table 4.

TABLE 4 - Summary of the occurrence of black-legged *Ochlerotatus*.

Aedes (*Ochlerotatus*)	Collections (1) of		Records (2) of		Dates of emergence (3)
	Larvae	Adults	Larvae	Adults	
cataphylla	early Apr. -early May	May 19	late Apr.	early May	May 14 -June 15
communis	early Apr. -early May	May 25 -June 22		mid-May	May 30 -June 7
hexodontus	early Apr. -early May	May 25 -June 22			
impiger				late May	May 19
implicatus	early Apr. -early June	May 19 -July 6			May 14 -June 17
intrudens	early Apr. -early June	June 2		June 5 -Aug. 18	
niphadopsis	early June				
pionips	early May		as late as mid-July		
punctor	early Apr. -early June	June 8 -Aug. 25	May		
spencerii	early Apr.		common in abundant late Apr. by May 10		

(1) Tables 2 and 3; (2) Rempel, 1953; (3) Klassen and Hocking, 1964.

It is apparent from the table that the larvae of most species appear very early in the season. However, *A. pionips* Dyar is perhaps a slightly later species, as indicated by Haufe (1952) and Rempel (1953), and it seems in *A. intrudens* and *A. punctor* that the hatching from eggs continues until later in the season, or the life span of the adults is longer. As for

A. niphadopsis it is not clear, as only one larva was collected.

For banded-legged mosquitoes of subgenus *Ochlerotatus*, similar data are also given in Table 5.

TABLE 5 – Summary of the occurrence of banded-legged *Ochlerotatus*

Aedes (*Ochlerotatus*)	Collections (1) of larvae	adults	Records (2) of larvae	adults	Dates (3) of emergence
campestris (4, 5)	late Apr.			early Jun.	May 19
canadensis	late Apr.			mid-May -late Jul.	May 30 -Jun. 7
dorsalis (4)	late May -late Jun.		generally early Jul.	late Jul. -Aug.	
excrucians (5)	early Apr. -early Jun.	Jun. 22	early May		May 27 -Jun. 4
fitchii·	early Apr. -early Jun.	Jun. 22 -Sept. 11	May	June -early Jul.	May 27 -Jun. 17
flavescens (4)	late May -early Jun.		mid-May	generally Jun. -Jul.	Jun. 7 -Jun. 17
increpitus	early Apr. -late May	Jun. 22 -Aug. 10	early May	late May -June	
riparius	early Apr. -early May	Jun. 22 -Aug. 10		mid-late May	
stimulans		Sept. 16	late May	late May -early Jul.	May 30 -Jun. 4

(1), (2), and (3) see table 4; (4) a second generation may occur; (5) long-lived species, occasional specimens may be encountered late in the season.

Of the species given in the table, *A. excrucians* Walker, *A. fitchii* (Felt and Young), *A. increpitus* Dyar, *A. riparius* Dyar and Knab, and *A. stimulans* (Walker) are considered woodland species and have only one generation a year. The larvae appear as early as most black-legged mosquitoes, but the emergence is delayed because of slower development, as indicated by Haufe (1953 and 1956) and as recognized by the fact that the black-legged mosquitoes emerged earlier than the banded-legged ones, when the larvae from the same pool were reared in the laboratory. The females were collected as late as September 11 in *A. fitchii*

as August 10 in *A. increpitus* and *A. riparius*, and as September 16 in *A. stimulans* . These facts seem to indicate that the life span of adults of those species is very long, as Rempel (1953) stated that occasional specimens of *A. excrucians* may be encountered in mid-summer. *A. canadensis* (Theobald) is also a wood-loving species. The larvae appeared as early as other banded-legged species mentioned above. Occasionally hatching occurs in the fall in Illinois (Horsfall, 1955).

Other tabulated species, *A. campestris* Dyar and Knab, *A. dorsalis* (Meigen), and *A. flavescens* (Müller), are grassland-lovers, and a second generation may occur, when the environment is favorable. They seem to be slightly later-appearing species than the woodland species.

Aedes vexans (Meigen), which belongs to the subgenus *Aedimorphus*, is found in the three main ecological zones in Saskatchewan, the prairies, aspen grove region, and coniferous forest (Rempel, 1953). This species seems to have multiple generations when the conditions are favorable. The larvae were collected in early May to late June and the adults on July 13 and August 10. It is apparently a late-appearing species.

Black-legged *A. (Aedes) cinereus* (Meigen) seems to.be rather late in appearance, though the first larva was collected in late April. The adults were collected from June 22 to August 25.

DISTRIBUTION PATTERN OF MOSQUITO LARVAE
Introduction

Populations of animals may be effectively estimated on the basis of their distribution pattern, and much has been published on this subject with various kinds of animals, among which however mosquitoes are not included. In applying the sequential sampling technique, which will be described later, and also in comparing the population densities at different pools, it is required to establish the nature of the frequency distribution pattern of mosquito larvae (and pupae).

A dipper is usually used for collecting mosquito larvae, and is considered a handy and reliable tool. Here, an attempt has been made to analyse the distribution pattern of mosquito larvae in their habitats by using the number per dip.

Collections Used for the Determination of the Frequency Distribution

Table 6 gives the data of collections of mosquito larvae for determining the frequency distribution pattern of the numbers per dip. Collections numbers 9 to 24 in the table are the same data as used for the seasonal fluctuation of *C. inornata* described earlier. The table indicates that the collections were made at various habitats of various sizes during the period covering May 25 to September 30, and the mosquito species collected were distributed in the genera *Anopheles*, *Culex*, *Culiseta*, and *Aedes*. The habitats included a grassland pool, a woodland pool, a collection of scattered small pools, and the marginal part of a creek, and the mosquitoes were found at some times as a single species, and at others mixed.

TABLE 6 - Collections of mosquitoes in immature stages for the frequency distribution pattern.

Collection number	Date	Habitat	No. of dips	Mosquitoes collected
1	May 30	Permanent pool in open place	100	*Anoph. earlei* ; *Aedes* spp.
2	May 30	Temporary grassland pool	100	*Anoph. earlei*
3	June 7	Collection of small pools in pasture	100	*Culiseta* spp.; *Aedes* spp.
4	June 21	Permanent pool in open place	50	*Anoph. earlei*
5	June 24	Marginal part of a creek	60	*Anoph. earlei*; *Culex territans*; *Culiseta inornata*
6	June 24	Same as No. 3	30	*Culiseta inornata*; *Aedes* spp.
7	July 29	Same as No. 3	100	*Culex territans*; *Culiseta* spp.
8	July 29	Same as No. 5	40	*Culex* spp.; *Culiseta inornata*
9 to 24	May 25 to Sept. 30	Permanent woodland pool (see page 189)	10-50	*Culiseta inornata*

The Relation Between Mean and Variance of the Numbers Per Dip

In Table 7 the mean, ($\bar{x}$), variance (s^2), and range of the numbers of mosquitoes per dip are given. The means vary from 0.02 to 39.40, and the variances from 0.02 to 3975.34. The minimum value of the range for most collections is zero, and the maximum value is up to 206. These figures indicate a great variability in number of mosquito larvae between the pools and also within each pool.

Several mathematical models have been developed to describe the distribution pattern of animal counts. When the distribution is considered random, a Poisson distribution is often applied. In the

Poisson distribution, the probability for a given positive integer x,
is given by $P(x) = e^{-m}m^x/x'$ . . . . (1)
where m is the mean. It is a property of the Poisson distribution
that the variance is equal to the mean, and the expression $\Sigma (x-\bar{x})^2/\bar{x}$
gives a good approximation to x^2 with (n-1) degrees of freedom,
where n is a sample size (Andrewartha, 1961).

TABLE 7 - Mean, variance, and range of the numbers of mosquitoes
          per dip, together with x^2 - test for significant departure
          from Poisson distribution.

Collection number	Mean (x)	Variance (s^2)	Range	x^2
1	1.61	5.47	0 - 11	336.60**
2	0.56	0.89	0 - 4	157.41**
3	0.49	2.76	0 - 15	557.37**
4	0.02	0.02	0 - 1	49.00
5	0.20	0.82	0 - 5	161.07**
6	14.60	509.21	0 - 85	1011.52**
7	1.00	2.51	0 - 10	248.49**
8	2.43	45.53	0 - 35	730.86**
9	0.02	0.02	0 - 1	49.00
10	15.00	561.11	0 - 76	336.69**
11	35.70	3975.34	0 - 206	1002.15**
12	33.30	1552.54	0 - 159	885.78**
13	32.40	882.04	0 - 82	244.98**
14	28.60	867.82	4 - 80	273.06**
15	39.40	2590.27	1 - 164	591.66**
16	17.40	703.38	0 - 88	363.78**
17	19.30	368.46	0 - 50	171.81**
18	11.40	212.93	1 - 44	168.12**
19	6.30	58.23	0 - 24	83.16**
20	5.50	74.28	0 - 26	121.59**
21	1.60	11.60	0 - 11	65.25**
22	0.45	1.52	0 - 4	64.22**
23	0.70	2.34	0 - 5	30.06**
24	0.20	0.18	0 - 1	8.10

x^2 with n-1 degrees of freedom is calculated by $(n-1)s^2/\bar{x}$. For
further explanation see text. For collection number see table 6.

**Discrepancy from Poisson distribution is significant at 1% level.

It is apparent from Table 7 that the variation of variance is much greater than that of mean, and the Poisson distribution does not seem to fit the data excepting collection numbers 4, 9, and 24. To make sure, the values of $\Sigma (x-\bar{x})^2/\bar{x} = (n-1)s^2/\bar{x}$ were calculated as the fifth column of Table 7; these were highly significant except for the above three collections. This shows that a random distribution - Poisson distribution - could not be rejected in the number of mosquitoes per dip, when the population density was as low as 0.20, and discrepancy from Poisson became greater with the increase of the mean, an aggregated type of distribution being indicated.

As stated by Waters (1959), there will be some field counts for which x^2 test will show no significant departure from either Poisson or an aggregated-type distribution such as negative binomial. It seems that non-significant values of x^2 in collections 4, 9 and 24 are attributable to the sparsity of the population and consequent low expectation of occurrence of mosquitoes in individual dips.

Goodness-of-Fit to the Poisson and the Negative Binomial

Insect counts in the field are often fitted fairly well by a negative binomial distribution (Andrewartha, 1961; Anscombe, 1949; Bliss, 1953), which is one of the aggregated-type distributions. The frequency distribution of the negative binomial is given by expanding the expression $(q-p)^{-k}$, where q-p = 1, p = m/k, m is mean, and k is a positive exponent. As the variance of a negative binomial approaches the mean, or the over-dispersion decreases, $k \rightarrow \infty$ and $p \rightarrow 0$. Under these conditions it can be shown that the distribution converges to that for the Poisson (Fisher et al., 1943).

Goodness-of-fit to the Poisson and the negative binomial was tested (Tables 8 to 11) for the data with 100 dips, i.e. collection numbers 1, 2, 3, and 7.

Theoretical frequencies for the Poisson were calculated successively by the following formulae. The probability of observing zero count, P(0), is

$$P(0) = e^{-m} \dots \dots \dots \dots \dots \dots \dots \dots \dots \dots \dots (2)$$

and the probability of observing (x+1), P(x+1), is

$$P(x+1) = m P(x)/(x+1), \dots \dots \dots \dots \dots \dots \dots \dots (3)$$

substituting sample mean, $\bar{x}$, for population mean, m. The theoretical frequency is obtained by multiplying each probability by the sample size, 100.

The formulae to be used for the theoretical values of the negative binomial (Bliss, 1953) are:

$$P(0) = (1+m/k)^{-k} \dots \dots \dots \dots \dots \dots \dots \dots \dots \dots (4)$$

and

$$P(x+1) = (x+k) m P(x)/ (x+1) (k+m) \dots \dots \dots \dots \dots \dots (5)$$

The constant k can be computed by a property of the negative binomial that the variance, σ^2, is equal to $(m+m^2/k)$, where m is mean, substituting again sample mean and variance, $\bar{x}$ and s^2, for m and σ^2.

In all of four examples shown in Tables 8 to 11, highly significant departure from the Poisson was demonstrated (p< 0.001),

which indicates that the distributions cannot be considered random. On the other hand, those distributions agree well with the negative binomial, except for collection 3, in which some discrepancy from the negative binomial is apparent. In this case, 15 larvae per dip were recorded once, which is a very high count compared with the others. This high count contributes larger variance, which in turn, yields rather small value of k responsible for the discrepancy. Generally speaking, the frequency distribution of the numbers of larvae per dip seems to agree with the negative binomial. The disagreement with the negative binomial in collection 3 may be attributable to sampling error.

TABLE 8 - Goodness-of-fit of Collection No. 1 to Poisson and negative binomial distributions.

No. of larvae per dip	Observed (O)	Frequency Hypothetical Poisson(P)	N. Binom. (N)	$\frac{(O-P)^2}{P}$	$\frac{(O-N)^2}{N}$
0	48	20.0	44.0	39.20	0.36
1	17	32.2	20.8	7.18	0.69
2	11	25.9	12.3	8.57	0.14
3	8	13.9	7.7	2.50	0.01
4	4 ⎤ 8				
5	4 ⎦	7.4	8.3	0.05	0.01
6	3 ⎤				
7	1				
8	1 ⎟ 8				
9	2	0.6	6.9	91.27	0.18
11+	1 ⎦				
Total	100	100.0	100.0	148.77	1.39

*P< 0.001; **0.50< P< 0.75   DF   4*   3**

Fitting the Negative Binomial Distribution with a Common k

Comparison between the means of two or more distributions are more direct and unequivocal if they have the same relative dispersion in terms of k, and two approaches to a common k were described by Bliss and Owen (1948). The first of them is a regression moment estimate applicable to the present data. The following calculation is based on Bliss and Owen (1958).

TABLE 9 - Goodness - of - fit of Collection No. 2 to Poisson and negative binomial distributions.

No. of larvae per dip	Frequency Observed (O)	Hypothetical Poisson(P)	N. Binom. (N)	$\dfrac{(O-P)^2}{P}$	$\dfrac{(O-N)^2}{N}$
0	66	57.1	64.6	1.39	0.03
1	20	32.0	22.5	4.50	0.28
2	8	9.0	8.2	0.11	0.00
3	4 ⎤				
4+	2 ⎦ 6	1.9	4.7	8.85	0.36
Total	100	100.0	100.0	14.85	0.67

*P< 0.001; **0.25< P< 0.50   DF   2*   1**

Two statistics, x' and y' are computed from the mean and variance of each component distribution:

$$x' = \bar{x}^2 - s^2/n \quad\text{.............................}\quad (6)$$
$$y' = s^2 - \bar{x} \quad\text{...............................}\quad (7)$$

where n is sample size. Their expectations are given exactly by

$$E(x') = m^2 \quad\text{...............................}\quad (8)$$
$$E(y') = m^2/k \quad\text{.............................}\quad (9)$$

Thus $(y'- x'/k)$ has zero expectation. For a single sample, we have the ratio

$$1/k_1 = y'/x' \quad\text{..............................}\quad (10)$$

as an estimate of $1/k$. The variance of $(y'-x'/k)$ is given to order $1/n^2$ by

$$V = 2m^2 (m-k)^2 [k(k-1) - (2k-1)/n - 3/n^2]/(n-1)k^4 \quad\text{...}\quad (11)$$

The invariance $w = 1/V$ is of the nature of a weight. If calculated by replacing m by $\bar{x}$, m^2 by x', and k by an empirical trial value of k', we can obtain an estimate of $1/k$, $1/k_c$, by

$$1/k_c = \Sigma (wx'y') / \Sigma (wx'^2) \quad\text{.................}\quad (12)$$

as the slope of a linear regression of y' on x', the regression line being constrained to pass through the origin (x' = 0, y' = 0).

Referring back to the data of Table 7, x' and y' were calculated by formulae (6) and (7) for each collection, and the relation between them is given in Fig. 2, in log scales so as to show the values with great variabilities in one chart.

Assumed that a proportional relation holds between the two, that is given by $y' = (1/k)x'$, then the relation is represented by a straight line with an inclination of one in the figure in log scales, because $\log y' = \log (1/k) + \log x'$. The data of Fig. 2 satisfies the

above assumption very well. This indicates that the relation between x' and y' is represented by a regression line passing through the origin, and, in turn the underlying frequency distributions are suggested to be the negative binomial with a common k.   It   is interesting that the same trend seems to be shown in the regression of y' on x' between collection numbers 1 to 8 for various species of mosquitoes and 9 to 24 for *C. inornata* (see Table 6), because the inclination of the regression line gives the estimate of k, which is considered an intrinsic property of the population sampled (Fisher et al., 1943).   However, it is likely that the value of k is species specific, and further studies are required.

It is known that in some cases k increases somewhat as m increases (Anscombe, 1949; Morris, 1954; Bliss and Owen, 1958). So, the values of $1/k_1$ calculated by equation (10) were plotted against mean, $\bar{x}$, in Fig. 3, which indicates, however, no appreciable relationship between the two. In order to know the exact situation, however, the number of dips for each collection seems to have not always been sufficient, and further investigations are required.

TABLE 10 - Goodness - of - fit of Collection No. 3 to Poisson and
            negative binomial distributions.

No. of larvae per dip	Frequency Observed (O)	Hypothetical Poisson(P)	N. Binom. (N)	$\dfrac{(O-P)^2}{P}$	$\dfrac{(O-N)^2}{N}$
0	77	61.3	83.3	4.02	0.48
1	15	30.0	7.2	7.50	8.45
2	4	7.3	3.3	1.49	0.15
3	1 ⎫				
4	2 ⎬ 4	1.4	6.2	4.83	0.78
15+	1 ⎭				
Total	100	100.0	100.0	17.84	9.86

*P< 0.001; **0.001< P< 0.005   DF   2*   1**

Now, a common value of k will be estimated. The statistics x' and y' for each of the distributions have already been obtained. The next step is to get an initial trial estimate of a common k, k'. As $\bar{x}$ varies excessively among the collections, a suitable equation for k' is

$$k' = g/\Sigma\ (y'/x') \quad \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots \quad (13)$$

where g is the number of collections. Thus we got $k' = 0.2822$. By using this value, $1/k_c$, an estimate of $1/k$, was obtained by equation (12) and as its reciprocal $k_c = 0.2947$, which does not differ so much from the first trial estimate $k' = 0.2822$. Thus we have estimated a common value of k at 0.2947. If k_c should differ appreciably from its trial value, k', recalculation is necessary by replacing the initial k' by k_c.

The required tests for agreement with a single k_c may be arranged as an analysis of variance:

Effect of	DF	SS	MS	F
Slope, $1/k_c$	1	B_0^2	B_0^2	B_0^2/S^2
Computed intercept against 0	1	$C+B^2-B_0^2$	I_0	I_0/S^2
Error	g-3	$[wy'^2]-B^2$	S^2	

where $B_0^2 = \Sigma^2(wx'y') / \Sigma(wx'^2)$
$[wx'^2] = \Sigma(wx'^2) - \Sigma^2(wx') / \Sigma w,$ $C = \Sigma^2(wy') / \Sigma w,$
$[wx'y'] = \Sigma(wx'y') - \Sigma(wx')\Sigma(wy')/\Sigma w,$ $B^2 = [wx'y']^2 / [wx'^2]$
$[wy'^2] = \Sigma(wy'^2) - C,$ $\Sigma^2(\text{---}) = (\Sigma(\text{---}))^2.$

TABLE 11 - Goodness-of-fit of Collection No. 7 to Poisson and negative binomial distributions.

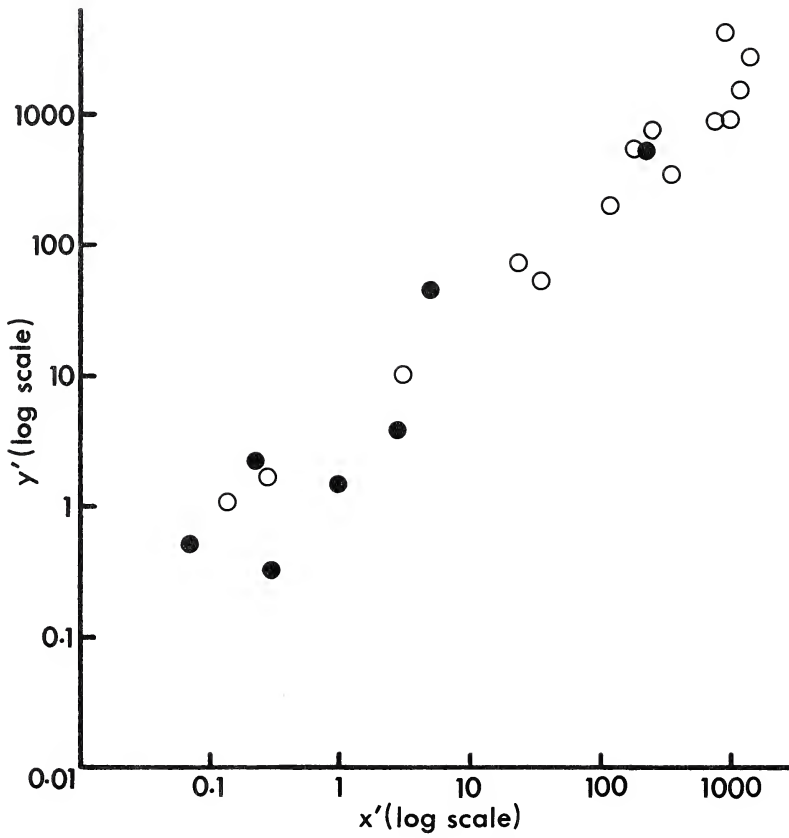| No. of larvae per dip	Observed (O)	Frequency		$\dfrac{(O-P)^2}{P}$	$\dfrac{(O-N)^2}{N}$
		Hypothetical			
		Poisson(P)	N. Binom. (N)		
0	53	36.8	54.4	7.13	0.04
1	21	36.8	21.7	6.78	0.02
2	16	18.4	10.8	0.31	2.50
3	4 ⎤				
4	1 ⎦ 5	7.7	9.0	0.95	1.78
5	3 ⎤				
6	1 ⎦ 5	0.3	4.1	5.39	0.20
10+	1 ⎦				
Total	100	100.0	100.0	20.56	4.54
DF				3*	2**

* P< 0.001; ** 0.10< P< 0.25

Fig. 2. Relation between two statistics, x' and y', defined by equations (6) and (7). ● : Collection Nos. 1 - 8; ○ : Collection Nos. 9 - 24. Collection Nos. 4 and 9 are not shown in the figure, because x' = 0, y' = 0, and also will be excluded in the later calculations, because of indeterminate values of y'/x'.

If a single k_c is justified, the F-value in the first row should be clearly significant and that in the second row not significant. The calculated values are shown below:

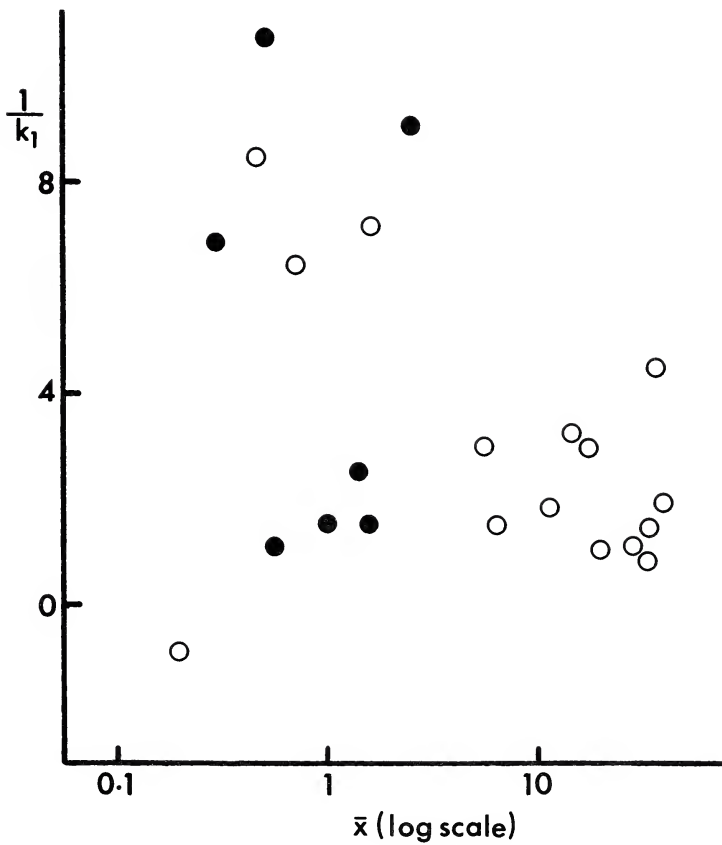| Effect of	DF	SS	MS	F
Slope, $1/k_c$	1	33.7809	33.7809	24.4670**
Computed intercept against 0	1	4.0297	4.0297	2.9186
Error	19	26.2327	1.3807	

Fig. 3. Relation between mean ($\bar{x}$) and estimate of $1/k$ ($1/k_1$). ● :
Collection Nos. 1 - 8; ○ : Collection Nos. 9 - 24.

The results are highly significant for slope and not significant for computed intercept against 0, that is a common value of k is justified.

Consideration of Reasons for a Negative Binomial Distribution

I have demonstrated that the number of mosquitoes per dip follows a negative binomial distribution with a common k. The negative binomial is generated by a distribution that is "contagious" in the sense that the presence of one individual in a division increases the chance of other individuals falling into that division. However, as Andrewartha (1961) stated, agreement with the negative binomial does not itself permit any inference about the biology of the mosquitoes, though a significant discrepancy from the Poisson series disproves the hypothesis of random scatter. In fact, according to Bliss (1953) the negative binomial may be regarded as being compounded from a number of Poisson series in which the means vary in such a way that they are distributed like x^2, and furthermore it is possible to imagine a number of other models to explain it.

The present data are not considered the sum of a number of Poisson series with different means, and other reasons should be sought.

One of them which might arise is a dipping error, however, its effect seems to be of little importance, or at least, the negative binomial distribution is not attributable only to it.

No habitat of mosquitoes in nature is considered so uniform that all parts of it are equally attractive to them. Marginal parts of a pool are usually preferable to mosquito larvae, and it is a common phenomenon that the spatial distribution of the mosquitoes is related to water-plants or overgrown vegetation. Thus the heterogeneity of the environment seems to be a great reason for the contagious distribution - the negative binomial. In fact, Hocking (1953) observed strong aggregation of the larvae of *Aedes communis* DeGeer , due apparently to the effect of sunlight and temperature gradient in the pool.

Another reason to be considered here may be a gregarious habit of mosquitoes. Although this has not been studied extensively, it seems important in the ecology of mosquitoes. It is commonly observed in the laboratory that mosquito larvae show some aggregated distribution in a tray, in which the environment does not appear to differ appreciably. This habit of aggregation differs in intensity with species, and, for example, strong aggregation of larvae is frequently seen in *Aedes aegypti* (L.), but it is hardly ever seen in *Anopheles hyrcanus sinensis* Wiedeman. The biological meaning of this is not clear at the present time, but is interesting in that it may be related to the level of optimum density of larvae. At any rate, the intrinsic behaviour of mosquitoes may play some role in the contagious distribution.

In short, the heterogeneity of habitat and possibly a sort of gregarious behaviour of mosquito larvae are considered to be responsible for the negative binomial distribution which is characterized by a larger variance than mean.

SEQUENTIAL SAMPLING TECHNIQUE

Introduction

Sequential sampling can be used for classifying a population into one of a number of pre-defined density levels, based on the accumulated results of each unit sampled. In classifying animal populations, it has been applied to the spruce budworm (Morris, 1954), whitefish, *Coregonus clupeaformis* (Mitchell) (Oakland, 1950), the lodgepole needle miner (Stark, 1952), and an aphid, *Myzus persicae* (Sulzer) (Sylvester and Cox, 1961). However, it has never been applied to mosquitoes.

The great value of this procedure lies in the fact that it involves a flexible sample size in contrast to conventional sampling procedures, and it would frequently be possible to determine whether or not a mosquito population requires control, or satisfactory control has been obtained, with the expenditure of much less time than would have been required if the number of sampling units was inflexibly fixed (Knight, 1964). Therefore, it would be reasonable to extend this technique to the immature stages of mosquitoes.

The procedure given by Morris (1954) is mainly followed by the present application.

Density Classes

As mentioned above, the sequential sampling technique is used for classifying a population into pre-defined density levels. It is desirable that density classes are determined so as to enable us to know from these classes whether or not the mosquito density is so high that control operations are necessary, or whether a control operation has been successful.

The density classes may be differently set up according to the situation in the city or town concerned. Here, I have classified density tentatively into three levels indicated by the critical mean number of larvae per dip as follows:

Density	Mean number of larvae per dip
Low	0. 1 or less
Moderate	Between 0. 5 and 2. 5
High	12. 5 or more

Density class "high" may be regarded as an indication that the mosquito density is so high that control is required, or that a control operation has influenced the population but little, and "low" may indicate that the density is so low that control is not required, or that control was satisfactorily done. "Moderate" is the intermediate situation between the two. Although the density is not so high control may be desirable if it is early in the mosquito season.

Of course, the necessity of controlling mosquitoes depends not only on the mosquito density in each habitat, but also on the relative area of the habitat compared with the whole area, as well

as the location of those habitats in relation to city or town to be protected from mosquitoes. However, it is still true that population density must be determined at each habitat before a decision to control is taken.

Acceptance and Rejection Lines

To apply the sequential sampling technique to the mosquitoes, of which number per dip is considered to follow the negative binomial distribution, it is necessary to find a common value of k fitting all the data with different levels of mean, and it has been determined as 0.2947.

The next step is to set up alternative hypotheses, H_0 and H_1, from the density classes. To distinguish between low and moderate densities at a certain probability level, H_0 and H_1 are that the number of larvae per dip is 0.1 or less and 0.5 or more, respectively; to distinguish moderate and high they are that the number is 2.5 or less and 12.5 or more. The values of the constants based on the negative binomial distribution at the critical densities under these hypotheses are shown in Table 12.

TABLE 12 - Values of the constants at the critical densities under the hypotheses of H_0 and H_1, based on the negative binomial distribution.

Constant	Density			
	Low - Moderate		Moderate - High	
	H_0	H_1	H_0	H_1
Mean = kp	0.1	0.5	2.5	12.5
p = kp/k	0.3393	1.6967	8.4833	42.4163
q = 1 + p	1.3393	2.6967	9.4833	43.4163
Variance = kpq	0.1339	1.3484	23.7083	542.7038

Each pair of hypotheses is accompanied by two possible errors: α and β are the probabilities of rejecting H_0 and H_1 at the respective critical densities. Here, both α and β were set at 0.10. A rather large value for error probability seems to be suitable for rapid mosquito survey, because it reduces the number of dips to be taken at each habitat and enables us to decide whether or not control is necessary by a quick evaluation of the population density over a wide area in a relatively short time.

Formulae for the acceptance and rejection lines then are:
$$d = sn + h_0 \quad \dots \dots \dots \dots \dots \dots \dots \dots \dots \dots \quad (13)$$
and
$$d = sn + h_1 \quad \dots \dots \dots \dots \dots \dots \dots \dots \dots \dots \quad (14)$$

where d is the cumulative number of larvae in the first n dips. The slope of the lines, s, is

$$s = k \log (q_1/q_0) / \log (p_1q_0/p_0q_1) \dots \dots \dots \dots (15)$$

where q_0 and q_1 are the values of q and p_0 and p_1 are those of p under the hypotheses of H_0 and H_1 (for actual figures see Table 12), and the intercepts of the equations (13) and (14) on the d-axis are

$$h_0 = \log B / \log (p_1q_0/p_0q_1) \dots \dots \dots \dots \dots \dots (16)$$

where $B = \beta / (1-\alpha) \dots \dots \dots \dots \dots \dots \dots \dots \dots \dots (17)$

and

$$h_1 = \log A / \log (p_1q_0/p_0q_1) \dots \dots \dots \dots \dots \dots (18)$$

where $A = (1 - \beta) / \alpha \dots \dots \dots \dots \dots \dots \dots \dots \dots \dots \dots (19)$

Thus we get the following formulae as acceptance and rejection lines for low versus moderate classes,

$$d = 0.2267n - 2.4153$$

and

$$d = 0.2267n + 2.4153,$$

and for moderate versus high

$$d = 5.0891n - 24.9138$$

and

$$d = 5.0891n + 24.9138,$$

as shown in Fig. 4. This graph may be used in the field to determine how many dips should be taken at each habitat in order to define the density class within the accepted limits of   and   . It is helpful to visualize each pair of lines as enclosing a band from which the plotted points must escape before the density class is satisfactorily defined.

For example, in collection number 1 mentioned earlier (see Tables 6 and 7), the first three dips show no larvae. When zero is plotted over each number of dips 1, 2, and 3, it is seen that they are within both bands of low-moderate and moderate-high. The fourth dip yields two larvae and the fifth none, therefore dips 4 and 5 are still within these bands. The sixth dip shows three larvae, so 2 + 3 = 5 is plotted over dip 6. This is shown to have escaped from the bands and to have fallen into the moderate zone, so dipping is discontinued. Thus collection number 1 is classified into moderate density. If the plotted points had escaped into the area above the higher band, the density would be classed as high, and if below the lower band, the density would be classed as low.

The Operating Characteristic Curves

The operating characteristic curves are useful aids in understanding how the plan operates. The curve is calculated from

$$L (p) = (A^h - 1)/(A^h - B^h) \dots \dots \dots \dots \dots \dots \dots (20)$$

$$p = [1 - (q_0/q_1)^h]/[(p_1q_0/p_0q_1)^h - 1] \dots \dots \dots \dots \dots (21)$$

where L(p) is the probability of accepting H_0 for any possible level of the population mean of kp, A and B are taken from equations (19) and (17), and h is a "dummy variable" which may be assigned convenient values.

The operating characteristic curve is shown in Fig. 5 by plotting L(p) against population mean, kp. The left-hand curve is for low versus moderate density classes. When the mean, kp, is 0.1, the probability of accepting H_0 (low density class) is 0.9;
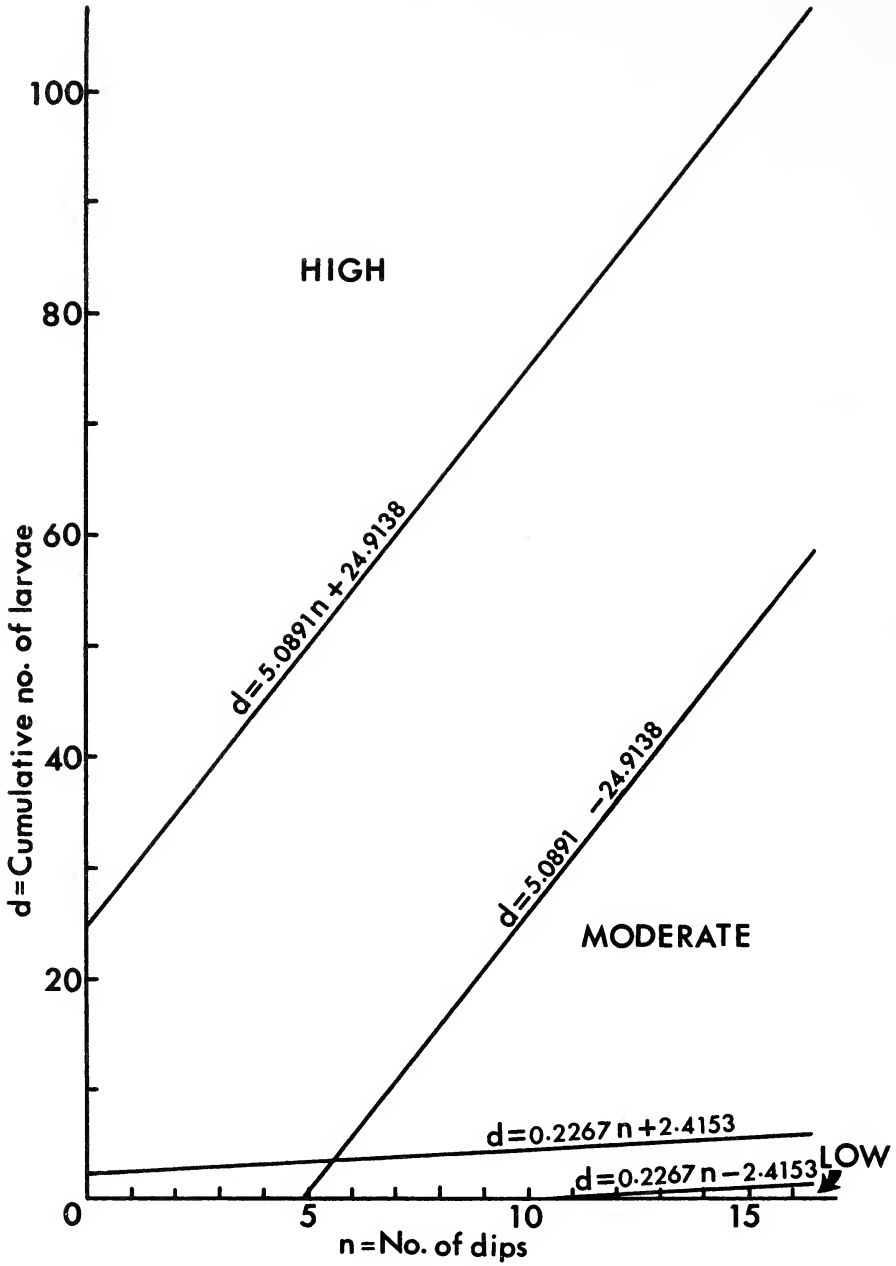
Fig. 4.   The acceptance and rejection lines.

accordingly the probability of accepting H_1 (moderate density class) is 0.1. When kp = 0.5 L(p) = 0.1 for H_0 and consequently 0.9 for H_1. At these two levels of kp, the probabilities correspond, of course, to those previously set for α and β. As kp decreases below 0.1, L(p) for H_0 becomes very low. When kp is ca. 0.23, the chances of accepting H_0 and H_1 are equal. The curve on the right is used in the same way for the moderate versus high density classes. The overlapping between the two curves is only at negligible probability levels. Thus the probability of considering a low density class high, or high density class low, is very small.

The Average Sample Number Curves

The average sample number curves can be drawn by plotting the values for E(n), the mean number of dips which must be taken, against kp, the mean number of larvae per dip, as shown in Fig. 6. For different values of kp, E(n) is calculated from

$$E(n) = [h_1 - (h_0 - h_1) L(p)] / (kp - s) \ldots \ldots \ldots \ldots \ldots (22)$$

where h_0, h_1, L(p), and s are taken from equations (16), (13), (20), and (15), respectively. E(n) does not indicate the number of dips which must be taken actually at each pool, but its expectation.

As would be expected, the peaks of the curves in Fig. 6 occur where populations are borderline between low and moderate or between moderate and high, which indicates that relatively more dips are required there.

Applications of the Sequential Sampling Technique in the Field

In applying the sequential sampling technique in the field it is convenient to use tabulations (Table 13) prepared from the acceptance and rejection lines, rather than the lines themselves. Dipping is continued until the cumulative number falls into one of the density classes. It is apparent from the table that at least 11 dips are necessary for the density to be classed into low, and at least six into moderate; if the number of larvae in the first dip is 31 or more, the density is classified as high without further dips.

Table 14 gives the results of applications of the sequential sampling technique to the data shown in Tables 6 and 7. It is demonstrated that the sequential plan can be used to classify the density correctly into one of low, moderate, and high density classes. The number of dips required for determining the class in various collections ranged from 1 to 20. When the density is high, the required number of dips was rather small, as expected from Fig. 6. This is of advantage in field work, because it takes much more time to count larvae dipped when the density is higher.

In sampling, the larvae are required to be dipped all over a larval habitat. In a large pool, dividing it into a few portions and applying the sequential plan at each will facilitate the work. Suggested larval survey form is given in Table 15.

This technique can be used effectively for the evaluation of the application of larvicides in a relatively short time. If the control operation is successful, then the densities at all pools will fall into
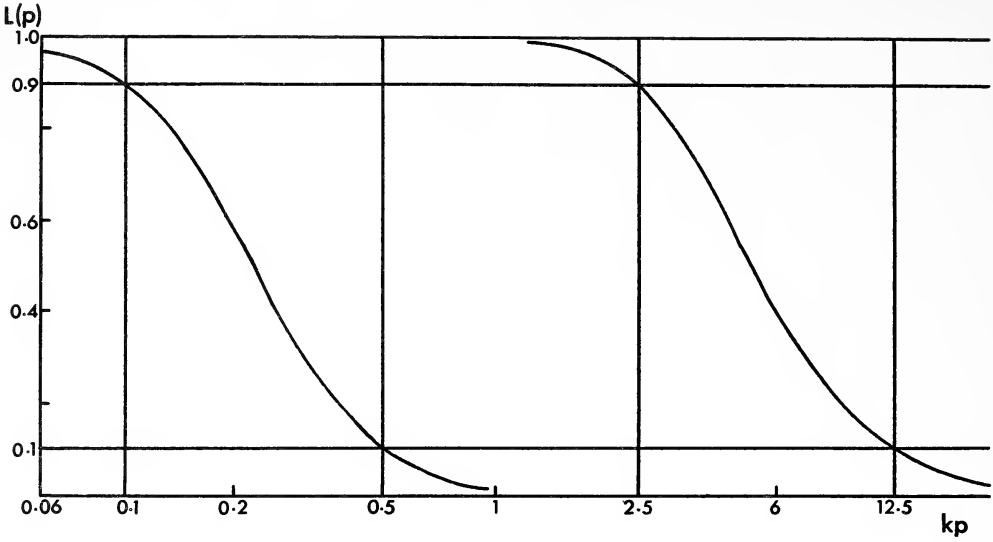
L(p)

Fig. 5. The operating characteristic curves for low versus moderate
         density classes (left) and for moderate versus high (right).
         kp = mean no. of larvae per dip; L (p) = probability of accept-
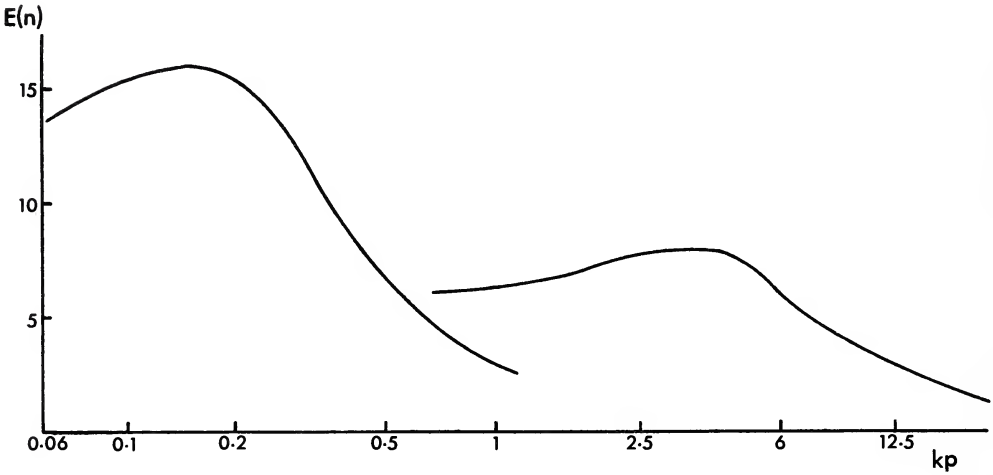   ing H_O hypothesis.

E(n)

Fig. 6. The average sample number curves for low versus moderate
         density classes (left) and for moderate and high (right).
         kp = mean no. of larvae per dip: E (n) = mean no. of dips to be
         taken.

the low density level. Also, this may be used for determining whether or not a second larvicide application is required specifically for the later-appearing mosquitoes. Necessity for mosquito control depends on the productivity of mosquitoes in a particular area, rather than the population density at each pool. To approach this, the following procedures may be appropriate. Firstly we determine the density class at each pool by the sequential sampling technique. Then, we take 0, 1, and 10 as indices for low, moderate, and high density levels, respectively, and multiply the index by the area of the pool (the area of the marginal parts if the larval distribution is confined there). If these are summed for a district to be examined, then it will represent the productivity of mosquitoes there. The sequential plan may be used for comparing regional differences of mosquito abundance, which provide us with the knowledge as to which region should be stressed for larval control operations.

TABLE 13 - Sequential table for use by field parties, prepared from the acceptance and rejection lines (Fig. 4).

No. of dips	Cumulative number of larvae		
	Low	Moderate	High
1			31 or more
2			36 or more
3			41 or more
4			46 or more
5			51 or more
6		4 to 5	56 or more
7		5 to 10	61 or more
8		5 to 15	66 or more
9		5 to 20	71 or more
10		5 to 25	76 or more
11	0	5 to 31	81 or more
12	0	6 to 36	86 or more
13	0	6 to 41	92 or more
14	0	6 to 46	97 or more
15	0	6 to 51	102 or more
16	0 to 1	7 to 56	107 or more
17	0 to 1	7 to 61	112 or more
18	0 to 1	7 to 66	117 or more
19	0 to 1	7 to 71	122 or more
20	0 to 2	7 to 76	127 or more

Continue to dip until the cumulative number falls into one of the 3 density classes of low, moderate and high.

TABLE 14 - Application of the sequential sampling technique to the
data shown in Tables 6 and 7 of Section 3.

Collection number	Mean no. of larvae	Density class determined	No. of dips required
1	1.61	Moderate	6
2	0.56	Moderate	20
3	0.49	Low	11
4	0.02	Low	16
5	0.30	Moderate	18
6	14.60	High	9
7	1.00	Moderate	20
8	2.43	Moderate	7
9	0.02	Low	11
10	15.00	High	1
11	35.70	High	2
12	33.30	High	2
13	32.40	High	2
14	28.60	High	1
15	39.40	High	1
16	17.40	High	7
17	19.30	High	3
18	11.40	High	3
19	6.30	High	2
20	5.50	Undetermined*	≥ 11
21	1.60	Moderate	9
22	0.45	Moderate	6
23	0.70	Moderate	7
24	0.20	Undetermined*	≥ 11

* During 10 dips made, the density class was not determined.

SUGGESTED STUDIES TOWARD
BETTER CONTROL OF EDMONTON MOSQUITOES

Introduction

In this section, only ecological questions are discussed,
although studies are also needed on the identification of mosquitoes
including the larvae in younger instars, the development of insec-
ticidal resistance, the methods and evaluation of applications of
chemicals, the effective and economical dosages of larvicides and
adulticides, the residual effects of insecticides when applied to the
habitat in the field, and so on.

TABLE 15 - Suggested mosquito larval survey form for the appli-
cation of sequential sampling technique in the field.

MOSQUITO LARVAL SURVEY FORM

Collection No.   Collector:
Place: Hour: , a.m. p.m.
Date: , 19
Breeding place
      - type: woodland - pool, grassland - pool, roadside - ditch,
           small pools in pasture, creek, other (   )
      - permanent, temporary
      - size
      - notes (marginal vegetation; water plants;   animals;
           temperature, pH, cleanness of water;   etc.)

No. of dips	No. of larvae	Cumula- tive no.	No. of dips	No. of larvae	Cumula- tive no.
1			11		
2			12		
3			13		
4			14		
5			15		
6			16		
7			17		
8			18		
9			19		
10			20		

Density class determined: Low, Moderate, High
Instar of larvae:
Species identified:

The Time of Hatching and Emergence

The prediction of the emergence time of mosquitoes is re-
quired to determine the appropriate time for chemical control. The
best time for controlling mosquito larvae is before they begin to
pupate, the pupae being much more resistant to insecticides than
larvae, but not before hatching is complete. Strictly speaking, the
above situation is hard to realize in the field, because the time of
hatching differs between species and also within species so that there
remain some eggs of *Aedes* to be hatched later in the season after
some adults have emerged. Thus the most effective time for insec-
ticidal applications against mosquito larvae is our special concern.
For this purpose, many points remain to be studied. These include

the studies on the time of oviposition and the durations of egg and
larval stages.

In mosquitoes belonging to the genera *Anopheles*, *Culex*, and
Culiseta, which overwinter as adults, the time of oviposition depends
on the time of blood feeding and the duration of egg development.
Blood feeding is certainly related to temperature and possibly to
adult diapause. The temperature apparently influences the matur-
ation of eggs.

All *Aedes* mosquitoes found around Edmonton overwinter as
eggs. According to Clements (1963), the different *Aedes* species fall
fairly clearly into those whose eggs enter diapause and require re-
activation, and those whose eggs merely become quiescent and hatch
shortly after exposure to an adequate hatching stimulus, although
they may require a few hours conditioning. Obligatory diapause in
the egg stage is found in *Aedes hexodontus* (Beckel, 1958), in *Aedes squamiger*
(Telford, 1958), and in *Aedes stimulans* (Horsfall and Fowler, 1961),
where exposure to low temperature is required before egg diapause
can be broken. These mosquitoes have only one generation a year.
Multivoltine species have facultative diapause, as in *Aedes dorsalis*
(Khelevin, 1958), *Aedes nigromaculis* (Telford, 1963), and *Aedes triseriatus*
(Baker, 1935), or have no diapause.

Most mosquitoes found around Edmonton have one generation
a year. However, there is a possibility that a second generation
occurs in some species, such as *A. campestris*, *A. dorsalis*, or
A. flavescens, perhaps in August when the conditions are favorable.

It is very likely that there is a wide variability in hatching
response of eggs, so that the time of hatching has a wide range, even
for eggs from the same batch.

Beckel (1958), Telford (1963) and others have discussed the
mechanism and ecological significance of egg diapause in mosquitoes,
and much has been published on the hatching response in quiescent
eggs of *Aedes aegypti* and some other *Aedes* species (see Telford, 1963).
However, the situation is still not clear for most *Aedes* mosquitoes.

After hatching from eggs, the development of larvae depends
on various factors. The most important are temperature, quality
and quantity of food, and larval density. It is expected that the
relation between larval period and temperature is described by an
equilateral hyperbola, or the relation between developmental speed
and effective temperature (temperature minus developmental zero
point) is linear, at least within a reasonable temperature range,
provided that other factors than temperature are constant. Based
on this relation, Haufe (1953) and Haufe and Burgess (1956) attempted
to predict dates of emergence in mosquitoes at Fort Churchill,
Manitoba, and stated: "The tundra species of mosquito (*A. impiger*
and *A. nigripes*) had lower thresholds of development approximating
34 F; the forest species (*A. communis*, *A. punctor*, *A. excrucians*)had a range
of 38 - 40 F, except *A. hexodontus*. The products of time and temper-
ature for the period of development of both tundra and forest species
were lower for the smaller than for the larger species". Studies of
this sort are desirable for all the mosquito species found abundantly

around Edmonton.

It is to be noted that the threshold of development obtained from the above relation is slightly higher than the actual value in the development of most insects, and is not necessarily the same as the critical temperature for hatching. Also, the developmental speed differs greatly according to factors such as quality and quantity of food and larval density. Thus, for the prediction of the date of emergence, careful investigations are required in the laboratory for each species. Another aspect to be involved is the relation between temperature in pools in various situations and meteorological records, for example see Haufe and Burgess (1956) and Haufe (1957).

Flight Range

Southwood (1962) stated: "It is suggested that animal movements fall basically into two types: trivial and migratory. Trivial movements are normally confined to the territory or habitat of the population to which the animal belongs, migratory movements carry the animal away from this area. Although there is undoubtedly no sharp line but a gradation between these two types, they can be distinguished by various ecological, physiological and behavioural characteristics", and "The ideal evidence of migratory movement is that while engaged in it the animal does not respond to food, a mate or habitat, and moves from the actual territory where it has developed into an inhospitable terrain: such movement is normally at the start of adult life". Provost (1957) reported the findings of a mark-and-release experiment with *Aedes taeniorhynchus* as follows: "Migration occurs the night of departure only, therefore twilight departures will result in longer migrations than middle-of-the-night departures. Appetential (trivial of Southwood, 1962) flights expand the range of occupation by a brood much beyond what is established by the migration." Thus mosquito dispersal consists of two phases of movement, and its range depends greatly on the migratory flight and to a lesser extent on the appetential flight.

In the appetential flight of mosquitoes, the distribution of breeding, resting, feeding, and oviposition sites, and in some species overwintering sites, will influence the degree of dispersal, because in mosquitoes these sites are situated quite often at different places and it is suggested that, within limits, the closer these are situated, the shorter the flight range. This should be considered in the field data, particularly when mark-and-release experiments are conducted.

As mentioned earlier, 14 species of adult female mosquitoes were collected around a pool near the University of Alberta. Excepting *Culiseta inornata*, all these mosquitoes are considered to have entered from outside or marginal parts of the City of Edmonton, since there are no breeding places for these species in the central part. This means that they dispersed at least a few miles, and should be considered potential pests for the Edmonton area according to their abundance. Here, it is required to determine the range of dispersal for each species. It is not known whether migratory flight

was involved in the dispersal or not. It seems reasonable to suppose that the range of dispersal is longer in mosquitoes where migratory flight is involved than in others. The investigation of this subject will help decide how widely insecticidal application or other control of larvae must be extended to control mosquitoes near Edmonton.

Fluctuation in Numbers

The habitats of most mosquito larvae are characterized by their unstableness. In years with small precipitation, the habitats will be greatly reduced in extent, though the amount of standing water is influenced by the dryness of the land to some degree, as indicated by Rempel (1953), and the reverse is also the case. The change of the habitats determines the area of breeding and oviposition places available for mosquitoes. Also, larval mortality has a close association with the amount of precipitation in some circumstances, since it is often observed that pools dry up before mosquitoes emerge. Thus mosquito abundance is expected to be closely correlated to the amount of precipitation. Temperature is also an important factor influencing mosquito populations, as indicated by Rempel (1953).

From the above statement, it is clear that there exists a close relationship between mosquito abundance and meteorological factors. However, the situation will differ from species to species, as evidenced by the fact that some species appear abundantly in one year and others in another year. The analysis of these correlations over a long period will help in studies on the population dynamics.

Another approach to studies on population dynamics is the analysis of mortality factors in the field, and also the influence of various environmental conditions such as trophic factors, population density, and so on, on the fecundity of adults in the field and also in the laboratory.

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EFFECT OF LARVAL DENSITY ON THE DEVELOPMENT
OF Aedes aegypti (L.) AND THE SIZE OF ADULTS*

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The effect of larval density of Aedes aegypti *(L.) on larval development and the size of resulting adults was studied in the laboratory. High larval mortality, long larval period, and small size of resulting adults were observed, when the larval density was high, as well as when the amount of food was small. Although the high larval density is often associated with shortage of food, it was demonstrated that even only the high larval density could produce these phenomena, when the amount of food per larva was kept constant. The effect of the density is considered to be expressed through increased stimulation of larvae by mutual contacts.*

INTRODUCTION

The effect of population density on the physiology and ecology of insects has received much attention by many investigators, as it is of basic importance in the study of population dynamics. As for mosquitoes, it is known that high larval densities are associated with high larval mortality, prolongation of the larval period, and small size of resulting adults with *Aedes aegypti* (L.) (Bar-Zeev, 1957; Shannon and Putnam, 1934), *Anopheles gambiae* Giles (Gillies and Shute, 1954), and *Anopheles quadrimaculatus* Say (Terzian and Stahler, 1949). Also Spielman (1957) and Krishnamurthy and Laven (1961) reported that overcrowding larvae of *Culex pipiens* L. f. *molestus* reduces the rate of autogeny among the resulting adults, and Gillies and Shute (1954) mentioned the change in maxillary index of *Anopheles gambiae* by larval overcrowding.

Although high larval density, or overcrowding, is often accompanied by a shortage of food, it seems to be advisable to separate the effect of density itself from that of starvation, since the two could be quite different processes. Shannon and Putnam (1934) seem to have made their experiments by increasing the larval density and keeping the food amount per container constant. If so, it is very likely that the larvae in high density were affected not only by the density itself, but also by the shortage of food. Bar-Zeev (1957) used a constant amount of food per larva in his experiments to demonstrate the effect of larval density, and said, "When the amount of food was not too high, and therefore, no film was formed, there was no undue mortality under crowded conditions; however, the development of the larvae was greatly delayed". This seems to have indicated the effect of density. However, he added "The growth rate was normal, provided that the amount of food per larva was

adequate, and that the water was renewed so as to prevent the development of a film of yeast. It can, therefore, be concluded that the inhibitory effect of crowded conditions on larval development is due to lack of food".

Thus it seems that no conclusion has been established for the effect of larval density itself in mosquitoes, and therefore, it was considered worthwhile to explore this further.

The effect of density would be investigated in an experiment with a constant quantity of food per individual at varying density levels (Klomp, 1964). On the other hand, if the quantity of food per container is kept constant, the larvae at high density will suffer shortage of food particularly in the latter part of development, as well as the effect of high density. In order to recognize the effect of food quantity free from the effect of density, food quantity would have to be changed at the same density level.

METHOD OF EXPERIMENTS

The mosquitoes used were *Aedes aegypti* kept at the Department of Entomology, University of Alberta. The eggs, not older than 15 days from oviposition, were allowed to hatch in water with a small quantity of dried yeast (Fleichmann's). The larvae which hatched within 12 hours were put into cups with 100 ml water containing dried yeast or rabbit pellets (North West Mill and Feed Co., Ltd.) or both. These cups were kept at constant temperatures, and the observations were made at a certain time every day. At each observation time, distilled water was added to keep a constant volume. When pupation occurred, the pupae were put into water in small glass vials with cotton plugs after recording their number, and emergence was awaited.

Four experiments were performed.

Experiment I— This was preliminary in nature. Density range was 1 to 64 larvae per cup, food used was yeast with quantity range of 1 to 64 units (1 unit = 1.7 mg) per cup, temperature, 25.7 ±1.5 C.

Experiment II— In this experiment, the quantity of food per cup was kept constant at various density levels. Density range was 1 to 128 larvae per cup, food used was 64 units of yeast plus 100 units of rabbit pellets per cup, temperature, 29.8 ± 1.2 C. From this experiment, the combined effect of food quantity and larval density will be seen.

Experiment III— This experiment was done to see the effect of different foods, that is 64 units yeast, 100 units rabbit pellets, 64 units yeast plus 100 units rabbit pellets, and 64 units yeast plus 200 units rabbit pellets. Density was kept constant at 16 larvae per cup, temperature, 29.8 ± 1.2 C.

Experiment IV – In this experiment, the quantity of yeast per larva was kept at 1 and 4 units, density range 1 to 256, temperature, 26.3 ± 0.9 C. Thus the effect of larval density will be seen from the data based on series of density levels at constant food quantity per larva. Also, by comparing in the same density level, the effect of food quantity will be demonstrated.

RESULTS OBTAINED

Effect of Larval Density on Larval and Pupal Mortalities

The larval and pupal mortalities in Experiments I, II, III, and IV are given in Tables 1, 2, 3, and 4, respectively.

In Experiment I, low larval mortality was observed at the density levels of 1 and 4 larvae per cup, when 4 to 64 units of yeast were supplied to each cup. With increasing density particularly when the amount of yeast was small, larval mortality became higher. No pupation occurred in the density 16 with 4 units of yeast per cup or in the density 64 with 4 or 16 units. No appreciable tendency was recognized in pupal mortality.

TABLE 1 - Mortalities of *Aedes aegypti* larvae and pupae reared at different densities with different amounts of yeast (Experiment I).

Density	Yeast (units)* per cup	larva	No. of repl.	Total no. of larvae	Larval mort. (%)	♂	♀	No. of pupae Total	Pupal mort. (%)
1	4	4	6	6	0.0	3	3	6	16.7
1	16	16	6	6	0.0	4	2	6	0.0
1	64	64	6	6	0.0	3	3	6	0.0
4	4	1	4	16	18.7	8	5	13	0.0
4	16	4	4	16	18.7	6	7	13	15.4
4	64	16	4	16	25.0	9	3	12	0.0
16	4	1/4	1	16	100.0	0	0	0	–
16	16	1	1	16	43.7	4	5	9	22.2
16	64	4	1	16	12.5	6	9	15	6.7
64	4	1/16	1	64	100.0	0	0	0	–
64	16	1/4	1	64	100.0	0	0	0	–
64	64	1	1	64	54.7	20	9	29	3.4

* 1 unit = 1.7 mg

Experiment II gave generally high pupation rate throughout the density levels of 1 to 128, indicating that the food used, 64 units yeast plus 100 units rabbit pellets, is suitable for larval survival. However, the larval mortality is lower at density 16 than at other densities, and this seems to indicate the optimum density for larval survival, with this combination of quantity and quality of the food.

TABLE 2 - Mortalities of *Aedes aegypti* larvae and pupae reared at different densities with a constant amount of food per cup (Experiment II).

Density	Replicates	Total no. of larvae	Larval mortality (%)	No. of pupae			Pupal mortality (%)
				♂	♀	Total	
1	23	23	13.0	12	8	20	5.0
4	12	48	6.2	21	24	45	4.4
16	5	80	1.2	43	36	79	1.3
64	3	192	3.6	100	85	185	1.6
128	2	256	16.4	126	88	214	0.9

Food used: 64 unit yeast plus 100 unit rabbit pellets per cup
(1 unit = 1.7 mg).

Experiment III, where the density of larvae was 16 per cup, shows that larval and pupal mortalities decrease from 64 units yeast to 64 units yeast plus 200 units rabbit pellets. This means that the lower food shown in the table is the better food for larval and pupal survival.

TABLE 3 - Mortalities of *Aedes aegypti* larvae and pupae reared with different foods (Experiment III).

Food used*	Replicates	Total no. of larvae	mortality (%)	No. of pupae			Pupal mort. (%)
				♂	♀	Total	
Y64	2	32	12.5	12	16	28	7.1
R100	2	32	9.4	15	14	29	3.4
Y64 + R100**	5	80	1.2	43	36	79	1.3
Y64 + R200	2	32	0.0	19	13	32	0.0

Density: 16 larvae per cup.
*Y: yeast; R: rabbit pellets; accompanied figure: quantity   per cup in units (1 unit = 1.7 mg).
**Data are from Table 2.

In Experiment IV, two series of the amount of yeast, that is 1 and 4 units per larva, were used. When the density was 16 or less, fairly high pupation was obtained, though the mortality is slightly higher with 1 unit yeast per larva than with 4 units. In density 64 with 1 unit yeast per larva, that is 64 units per cup, larval mortality was more than 50%, and only males pupated. In the density of 256 with 1 unit yeast per larva, that is 256 units per cup, larval mortality further increased up to 87%, 40% of pupae failed to emerge, and very low proportion of females was obtained. Very high larval mortality was observed also in the density of 256 with 4 units of yeast per larva, that is 1024 units per cup. This amount of yeast seemed to be too much for 100 ml water, because a film was formed on the water surface and high mortality occurred in earlier instars, unlike other combinations of density and food amount. Thus such a very low pupation rate as 2.9% is not due to the effect of high larval density, but probably to the film formation or other unfavorable conditions of the culture medium.

TABLE 4 - Mortalities of *Aedes aegypti* larvae and pupae reared at different densities with 2 series of a constant amount of yeast per larva (Experiment IV).

Density	Yeast (units)* per cup	larva	No. of repl.	Total no. of larvae	Larval mort. (%)	No. of pupae ♂	♀	Total	Pupal mort. (%)
1	1	1	32	32	9.4	18	11	29	3.4
4	4	1	13	52	20.8	23	19	42	7.1
16	16	1	5	80	13.7	44	25	69	1.4
64	64	1	2	128	57.8	54	0	54	1.9
256	256	1	2	512	86.9	64	3	67	40.3
1	4	4	32	32	12.5	20	8	28	3.6
4	16	4	13	52	0.0	27	25	52	0.0
16	64	4	5	80	13.7	42	27	69	2.9
64	256	4	2	128	23.4	60	38	98	5.1
256	1024	4	2	512	97.1	8	7	15	0.0

* 1 unit = 1.7 mg

In short, larval mortality generally increases with increased density and decreased food quantity through the shortage of food and the larval density itself. There seems to be an optimum density for larval survival, which differs from the minimum density. If the conditions are not suitable, then the favored sex is the male.

Effect of Larval Density on Pupation Curve

Frequency curves of pupation by sex in four experiments are shown in Figs. 1, 2, 3, 4, and 5. Males pupated earlier than females throughout the experiments. Generally a shorter larval period is seen in the cups where the density is lower and the amount of yeast is larger. When larval periods are compared on the basis of the same density with different amounts of food (see Fig. 1; compare Figs. 4 and 5), a longer larval period is seen with the decreased amount of food.

When the amount of food per larva was kept constant and the density of larvae was increased, the delay in development is clear, as seen in Experiment IV (Figs. 4 and 5). This is attributable to the effect of high larval density, not to the shortage of food, because the comparisons were made on the basis of the same amount of food per larva.

Here, it is apparent that the larval development is affected not only by the quantity of food, but also directly by the larval density, and the effect is more remarkable, when the amount of food per larva is smaller.

It is interesting that the longer larval period is usually associated with increased variation in larval period and with a tendency to be skewed towards the right. If a pupation curve is normally distributed, then it is expected that a cumulative percentage frequency of pupation in probit will be linear. Now, the normality of the pupation curves in Experiments II and IV, in which a fairly large number of larvae was used, was examined.

Cumulative percentage pupation in probit is plotted against larval period (days) in Figs. 6 to 11. When the density is low and food amount is large a linear relation is seen, that is, those pupation curves are shown to follow the normal distribution. The deviation from the normal distribution becomes remarkable with increasing density and decreasing food quantity. Thus there is some deviation from the normal distribution in the pupation curve, particularly when the conditions are unfavorable for larval development. Even when conditions are good, a few individuals sometimes pupate very late. For this reason, it seems that the median is a better representative of larval period than the mean.

Effect of Larval Density on Larval and Pupal Periods

Figs. 12, 13, and 14 show the relation between median larval period and larval density per cup for Experiment I, II, and IV, respectively. In these figures, the points with the same amount of food per cup were connected by straight lines. Generally, the median larval period becomes longer with increasing larval density. This is rather natural, because the amount of food per larva decreases with increasing density.

By connecting the points with the same amount of food per larva, the data for Experiments I and IV are represented in Figs. 15 and 16. In density levels of 256 and 64 of Experiment IV, a longer median period was obtained than in 1, 4 or 16, in spite of the
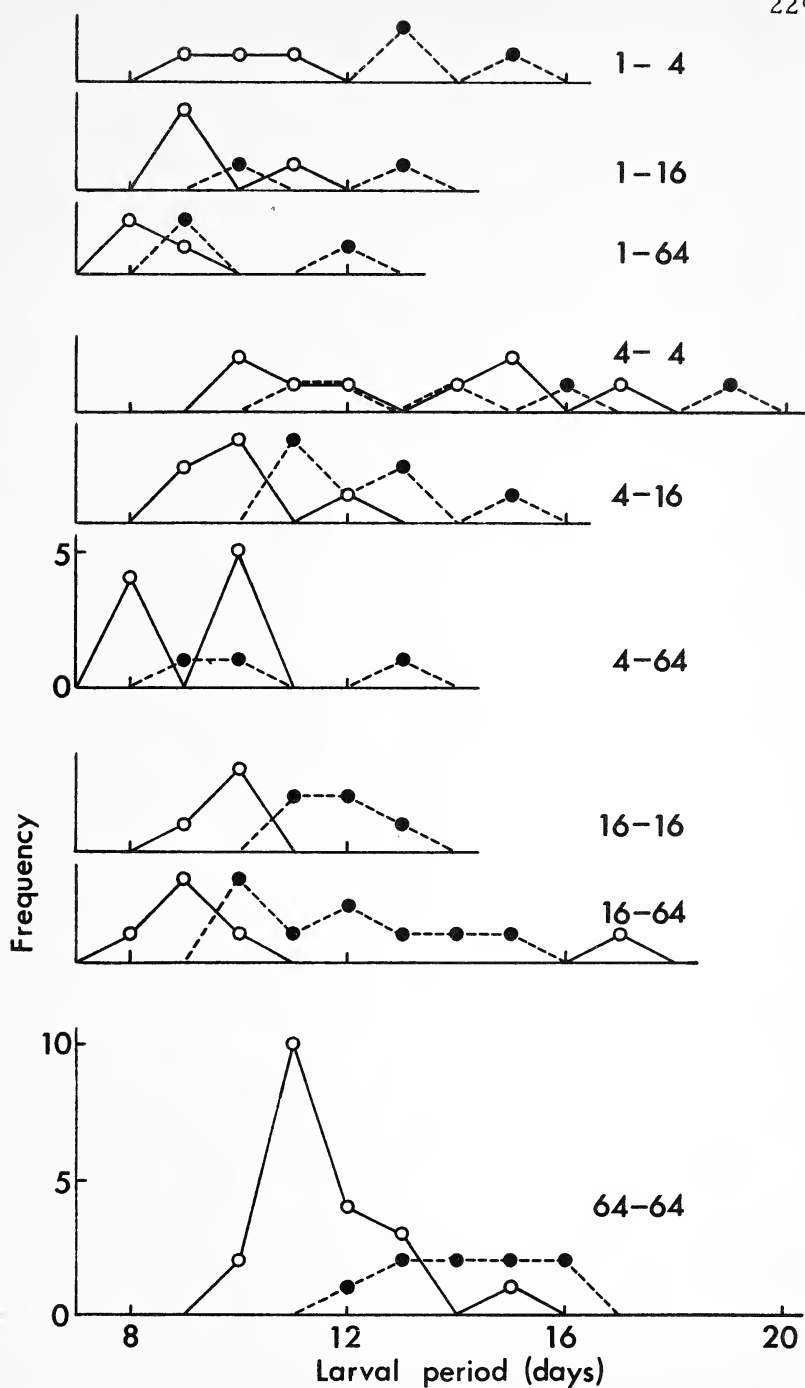
Fig. 1. Frequency distributions of larval period of *Aedes aegypti* (Experiment I). 4-64, for example, indicates that the larval density is 4 and the amount of yeast is 64 units per cup. ○: males; ●: females.
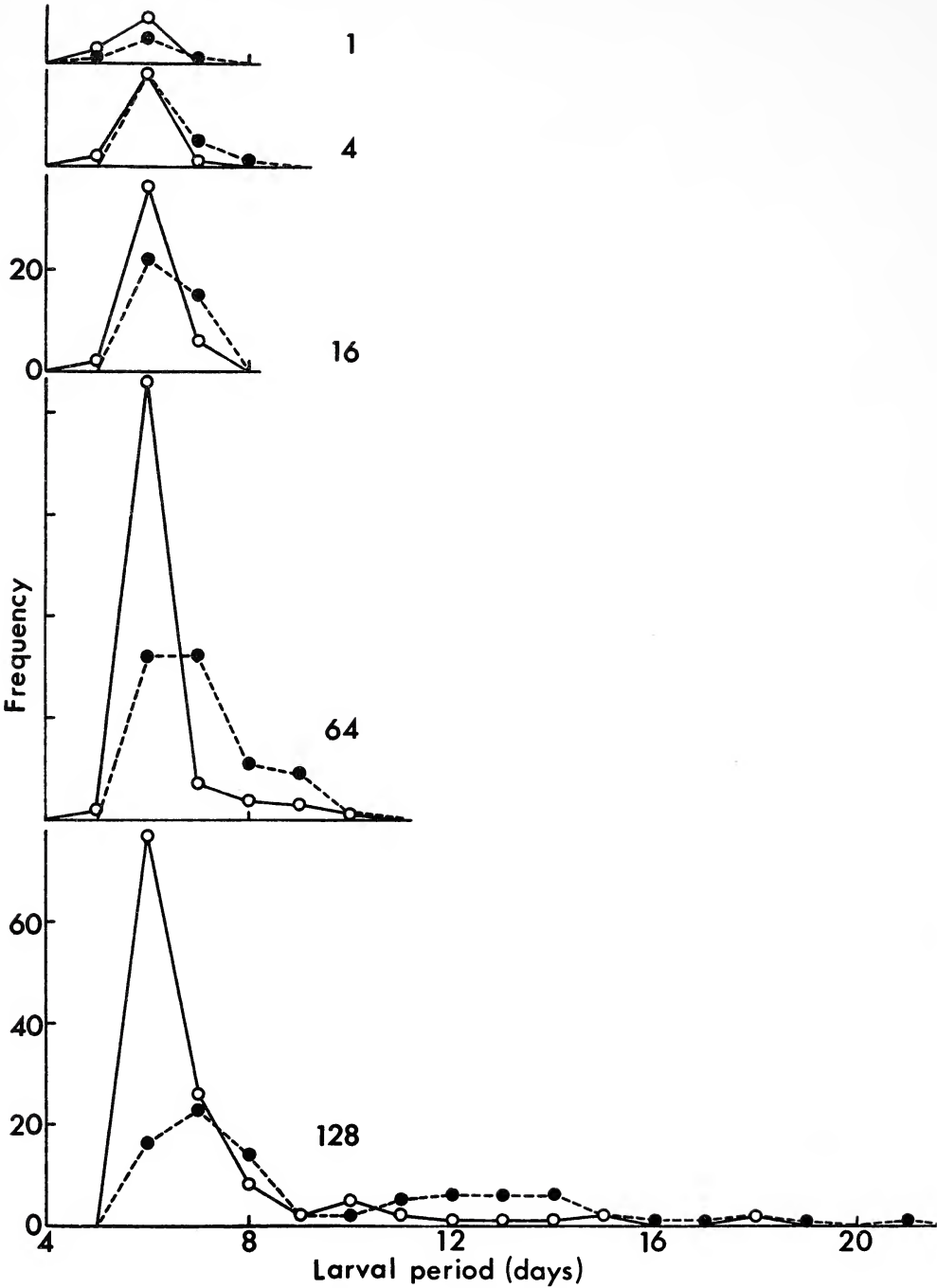
Fig. 2. Frequency distributions of larval period of *Aedes aegypti* (Experiment II). Figure shown indicates larval density. ○ : males; ● : females.

fact that the amount of food available for each larva in higher densities is the same as, or even slightly larger than, in lower densities. Here, the effect of high larval density is again suggested. Also in Experiment I, the tendency of the median to increase is seen at the density levels of 64 or more. It is interesting that there seems to exist a valley in median larval period at density 16, particularly
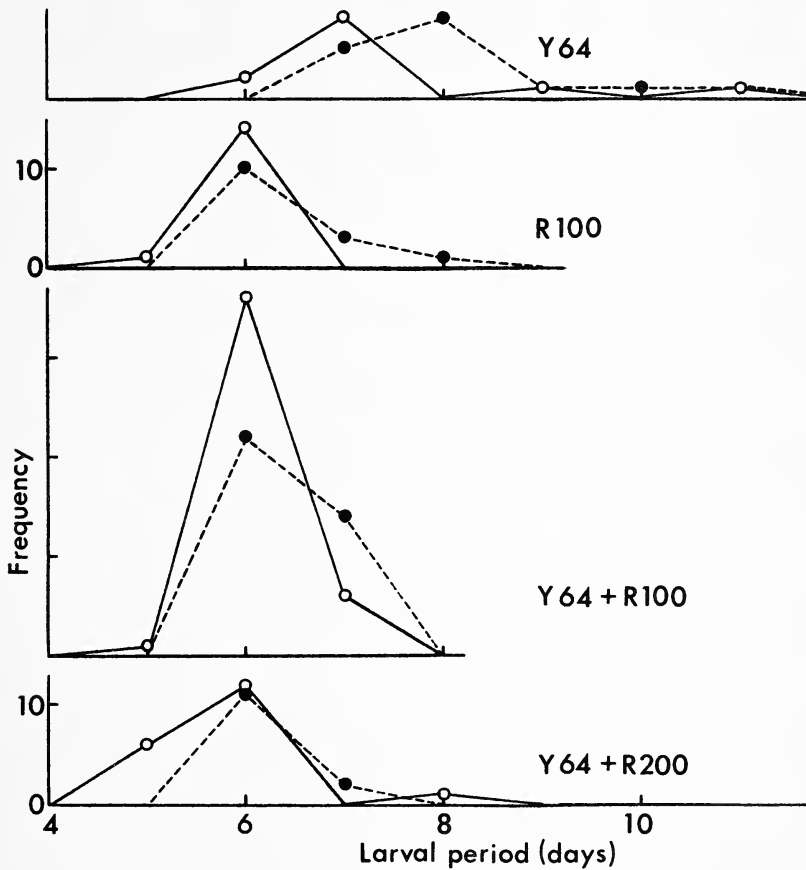
Fig. 3. Frequency distributions of larval period of *Aedes aegypti* (Experiment III). Y and R and accompanied figure indicate yeast and rabbit pellets and their amount in units. O : males, ● : females.
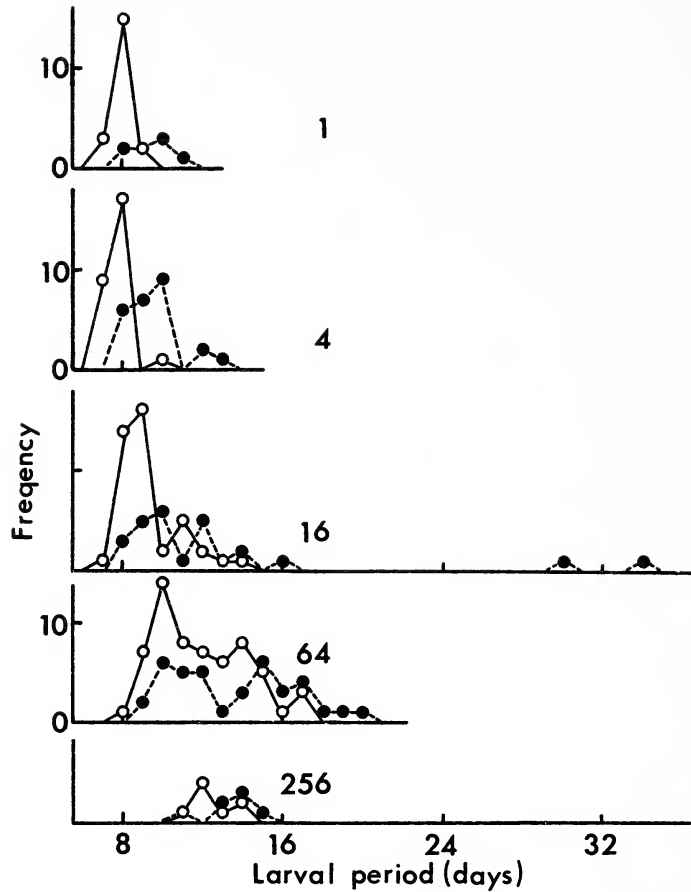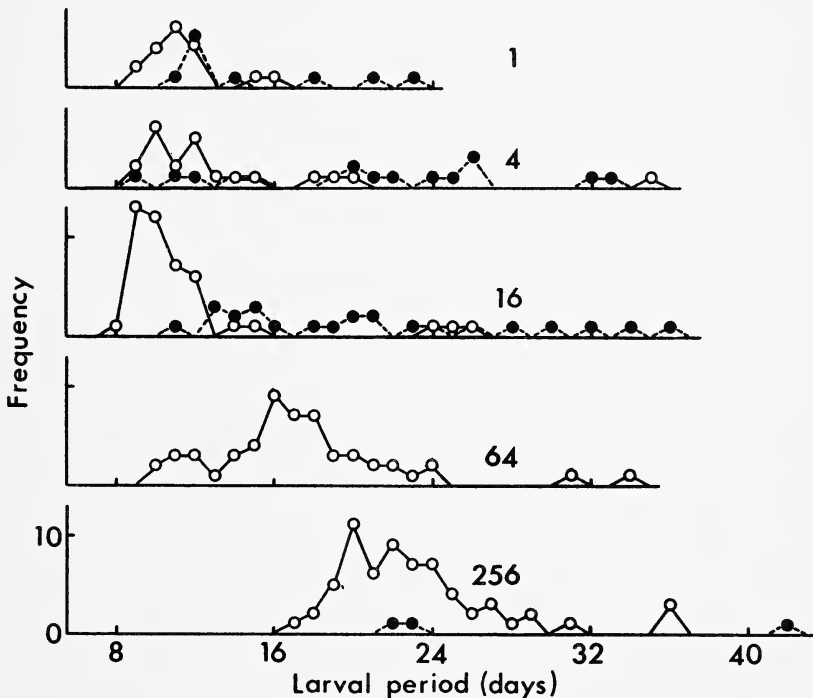
Fig. 4. Frequency distributions of larval period of *Aedes aegypti* (Amount of yeast per larva: 4 units; Experiment IV). Figure shown indicates larval density. ○ : males; ● : females.

when the amount of food is small, and furthermore, in the food amount of 1 in Experiment IV, the median becomes again smaller at density 1 than 4. The reasons for such peculiarities of the curves are not clear, but it seems that the median is determined by a balance between the effects of larval density and the amount of food available, and perhaps some other factors.

No distinct difference in pupal period was recognized among various amounts of food nor among larval density levels, though pupal density may affect the period. It seems that the pupal period is affected only by temperature, or at least, if some other factors affect it, their effect is very small. In Table 5, mean pupal periods in days are given by sex at the three different temperatures. The female has a slightly longer pupal period than the male.

It would be practically right to suppose that the larval period is determined by temperature, larval density, and the conditions of culture medium such as the quality and quantity of food, but the pupal period is determined only by temperature. The ratio of larval period seems, then, to indicate the suitability of the conditions for larval development. This ratio may be used to compare the larval period, even when experiments were made at different temperatures.

The calculated values for the ratio are shown in Tables 6 and 7, and compared on the basis of the same combinations of larval density and food amount in different experiments. The ratios for the combinations of D1Y4 (density 1 larva per cup, yeast 4 units per cup), D4Y16, and D16Y64 agree quite well among experiments, but those for D4Y4, D16Y16, D64Y16, and D64Y64, are rather

Fig. 5. Frequency distributions of larval period of *Aedes aegypti* (Amount. of yeast per larva: 1 unit; Experiment IV).   Figure shown indicates larval density.   O : males;   ● : females.

different from one another. The number of larvae used in Experiment I was not sufficient, and the latter combinations are considered somewhat unsuitable so that very slight differences in the conditions will make rather great changes in larval development. These would be responsible for rather great difference of the ratios in the latter group of combinations.

The above procedure will be valid only if the ratio of larval period to pupal period is constant over a reasonable temperature range. For this reason, further studies are required to determine the usefulness of the ratio. However, it is clear from the tables that larval period varies greatly with the quantity and quality of food at the same density level, and also that the same amount of food per cup, or even per larva, does not give the same larval period at different density levels. Therefore, care should be taken in attempting to determine the larval period at a certain temperature, or the developmental zero of mosquito larvae by rearing them at different temperatures.
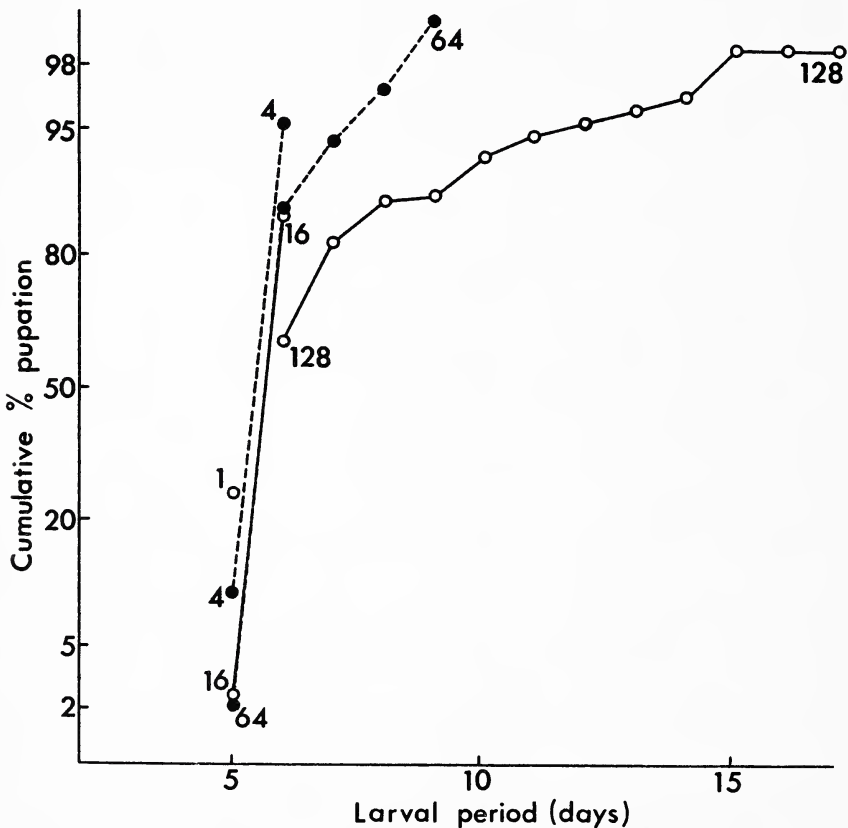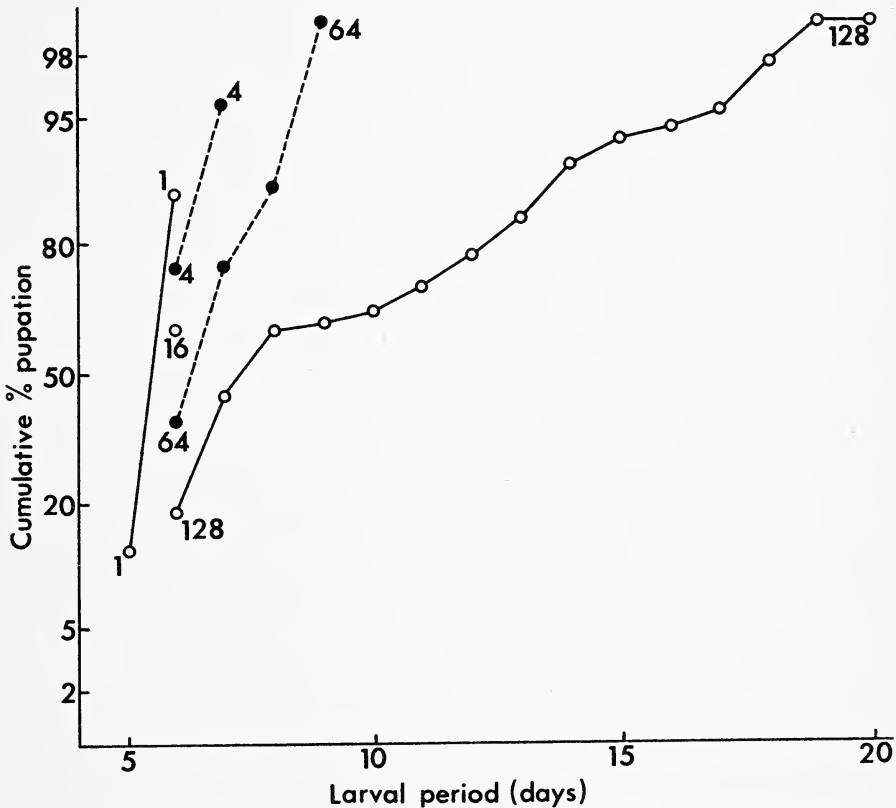

Fig. 6. The relation between cumulative percentage pupation (probit scale) and larval period in males of *Aedes aegypti* (Experiment II). Figure shown indicates larval density.

Effect of Larval Density on Body Size of Resulting Adults

In Figs. 17 and 18, the frequency distributions of wing length of the resulting adults in Experiments II and IV are given.

In Experiment II (Fig. 17), the wing length increases in both sexes slightly from density 1 to 16 larvae per cup, and decreases greatly with increasing density from 16. Fig. 18 shows the similar situation in Experiment IV, except for density 256 with yeast 4 units per larva, where the wing length is not considered to reflect the effect of this density, owing to high larval mortality in the earlier instars, as mentioned earlier. However, the changes in wing length are less remarkable than in Experiment II. This is due to the fact that the quantity of food per cup was kept constant in Experiment II, on the other hand in Experiment IV the quantity per larva was kept constant. Nevertheless, the apparent effect of larval density on the wing length can be seen in Experiment IV (Fig. 18).
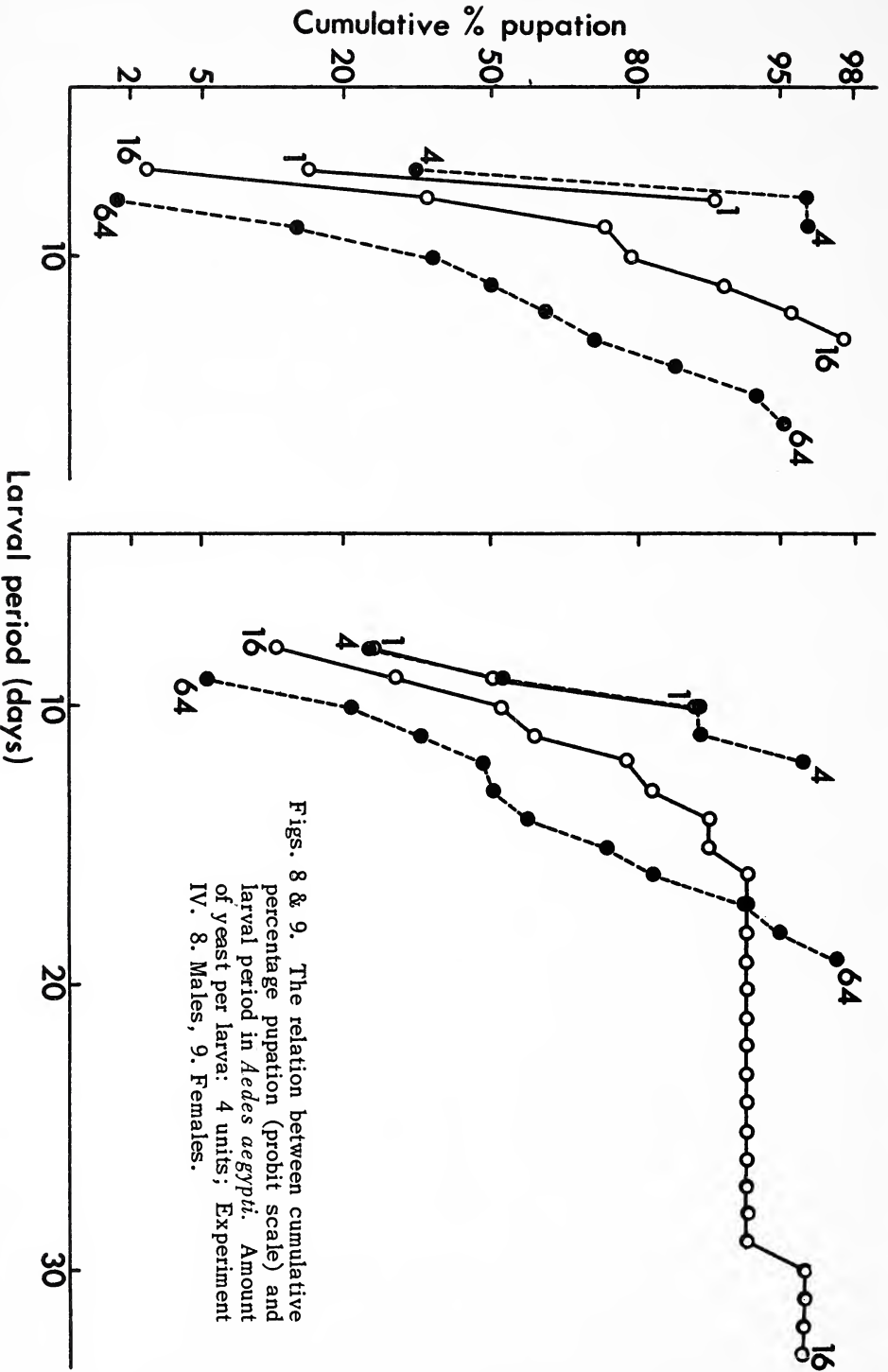

Fig. 7. The relation between cumulative percentage pupation (probit scale) and larval period in females of *Aedes aegypti* (Experiment II). Figure shown indicates larval density.

Figs. 8 & 9. The relation between cumulative percentage pupation (probit scale) and larval period in *Aedes aegypti*. Amount of yeast per larva: 4 units; Experiment IV. 8. Males, 9. Females.

It seems that the wing length of females is more sensitively affected than that of males with decreasing suitability for the larval stage, so that considerable overlapping in wing length of both sexes appears, as for example between densities 64 and 128 in Experiment II (Fig. 17). When the conditions become still less suitable, only males will pupate, as indicated from the densities 64 and 256 with yeast 1 unit per larva.

It is interesting that the frequency curve becomes steeper at the right hand side with decreasing suitability in the conditions for larval development, but the reasons for this are not yet clear.

Figs. 19 and 20 show the frequency curves of thorax length in Experiments II and IV. The thorax length shows a similar tendency to the wing length, excepting that the steepness of the curves at the right hand side is not seen, when the conditions become unfavorable.
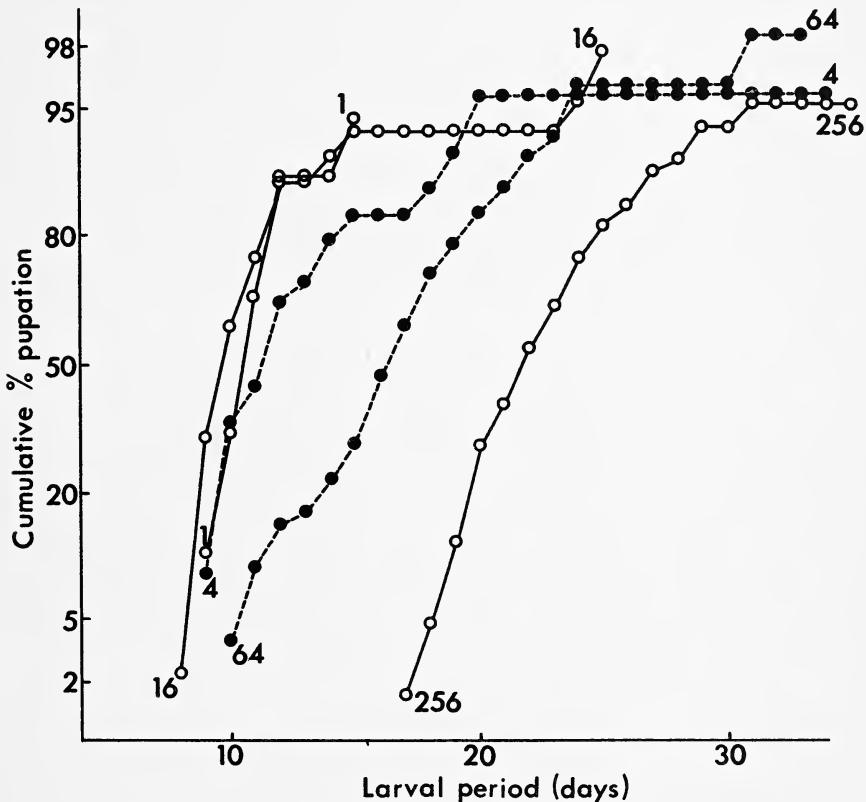

Fig. 10. The relation between cumulative percentage pupation (probit scale) and larval period in males of *Aedes aegypti* (Amount of yeast per larva: 1 unit; Experiment IV). Figure shown indicates larval density.
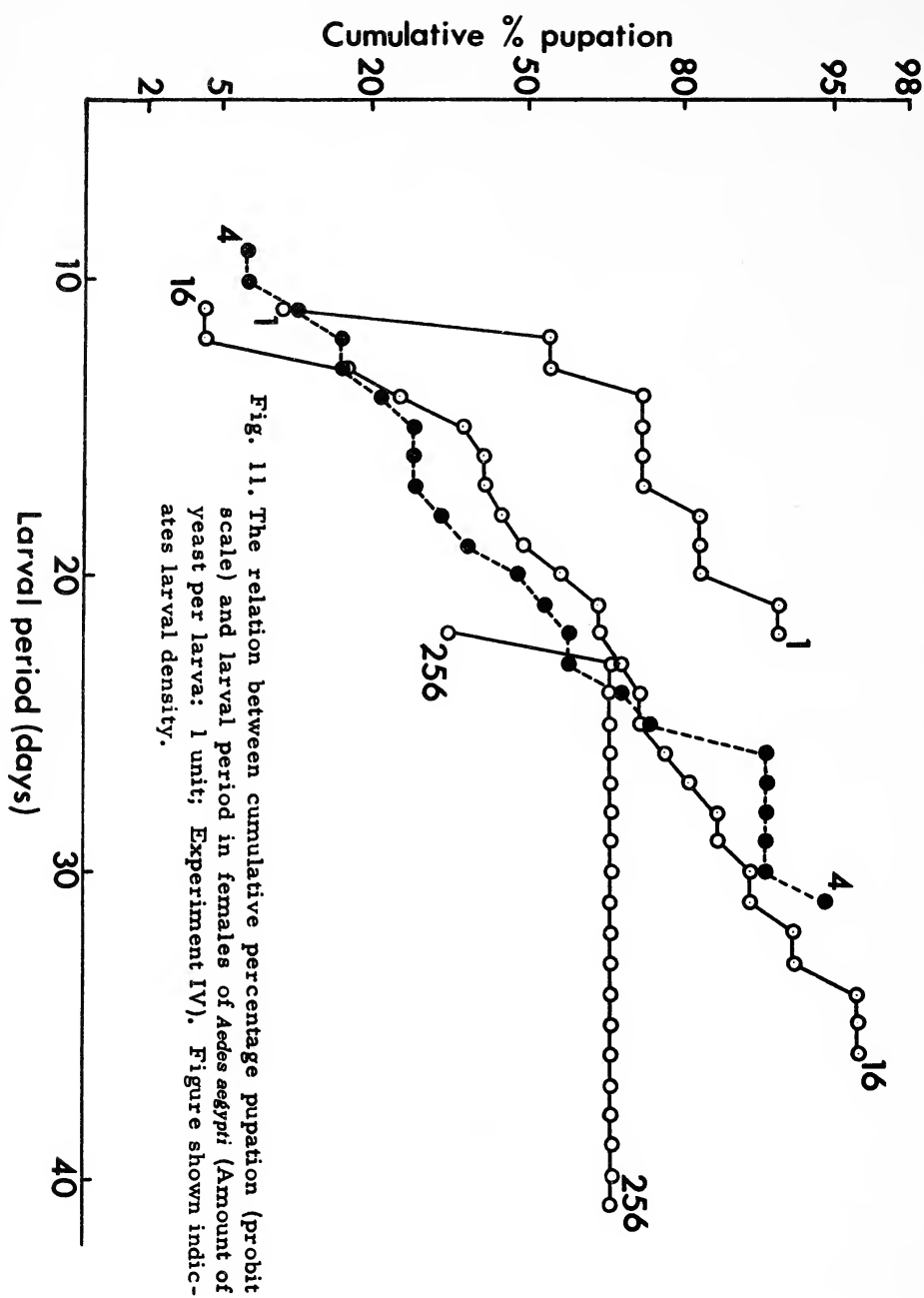
Fig. 11. The relation between cumulative percentage pupation (probit scale) and larval period in females of *Aedes aegypti* (Amount of yeast per larva: 1 unit; Experiment IV). Figure shown indicates larval density.

TABLE 5 - Pupal periods of *Aedes aegypti* by sex at different temperatures.

Experiment	Temperature C	Mean pupal period (days)	
		Male	Female
I	25.7	2.76	2.78
II and III	29.8	1.83	1.94
IV	26.3	2.36	2.42

TABLE 6 - The ratio of larval to pupal period of the males of *Aedes aegypti* (Experiments I - IV).

| Density	Food used*	Exp. I	Exp. II	Exp. III	Exp. IV	Mean
1	Y1				4.45	4.45
	Y4	3.44			3.14	3.28
	Y16	3.15				3.15
	Y64	2.83				2.83
	Y64 + R100		2.92			2.92
4	Y4	4.53			4.79	4.66
	Y16	3.37			3.05	3.21
	Y64	3.08				3.08
	Y64 + R100		3.01			3.01
16	Y4	**				**
	Y16	3.37			4.07	3.72
	Y64	3.15		3.55	3.56	3.42
	R100			2.98		2.98
	Y64 + R100		3.07			3.07
	Y64 + R200			2.89		2.89
64	Y4	**				**
	Y16	**				**
	Y64	3.91			6.86	5.39
	Y256				4.66	4.66
	Y64 + R100		3.07			3.07
128	Y64 + R100		3.19			3.19
256	Y256				9.24	9.24
	Y1024				5.00***	5.00***

* See Table 3.
** Unable to pupate.
*** Film was formed on water surface, larval mortality was very high.
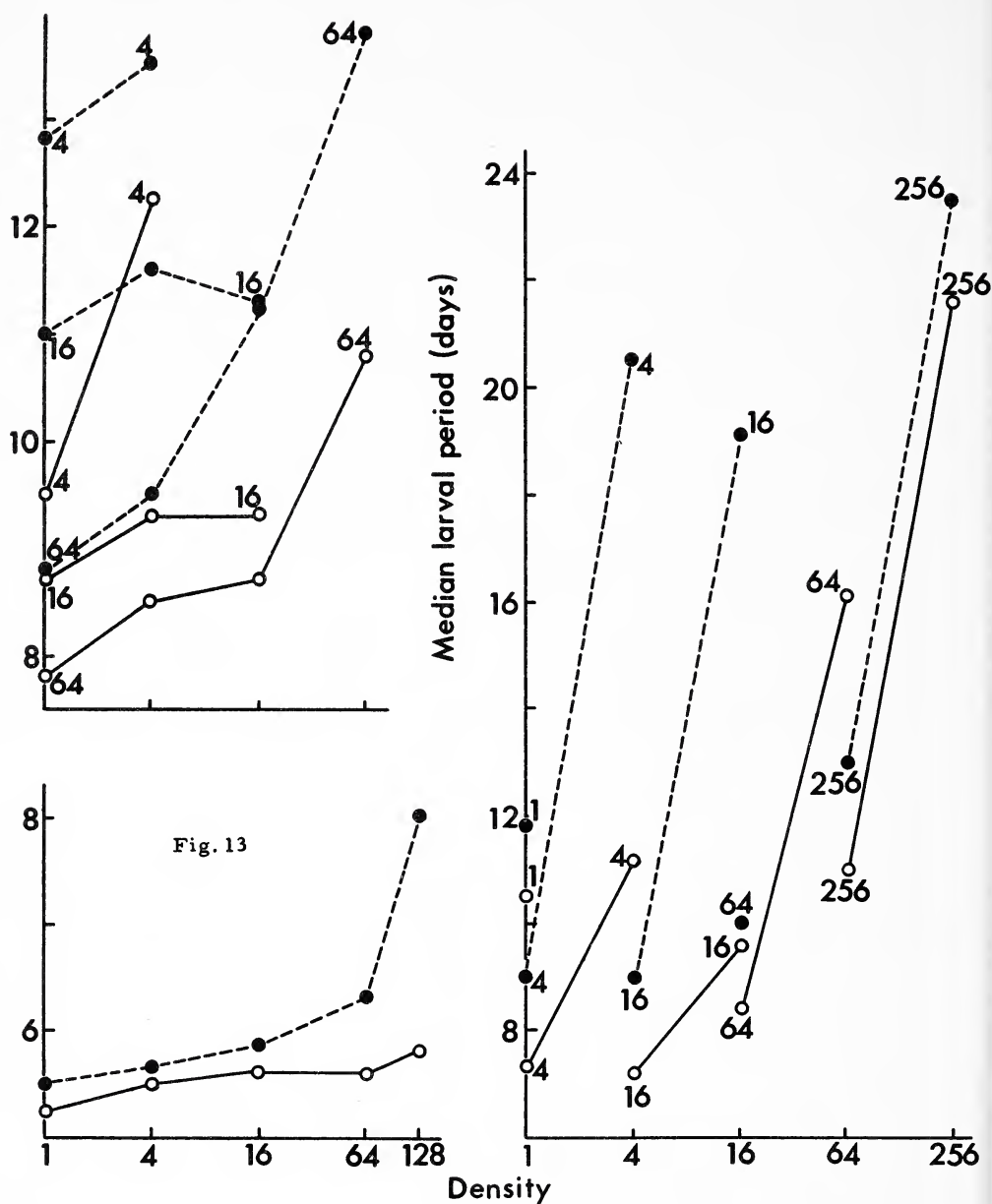
Figs. 12-14. Median larval period of *Aedes aegypti* at each density level.
Points for the same amount of yeast per cup are connected by
lines. 12. Experiment I. 13. Experiment II. 14. Experiment IV. Figure shown indicates the units of yeast per cup.
O : males; ● : females.
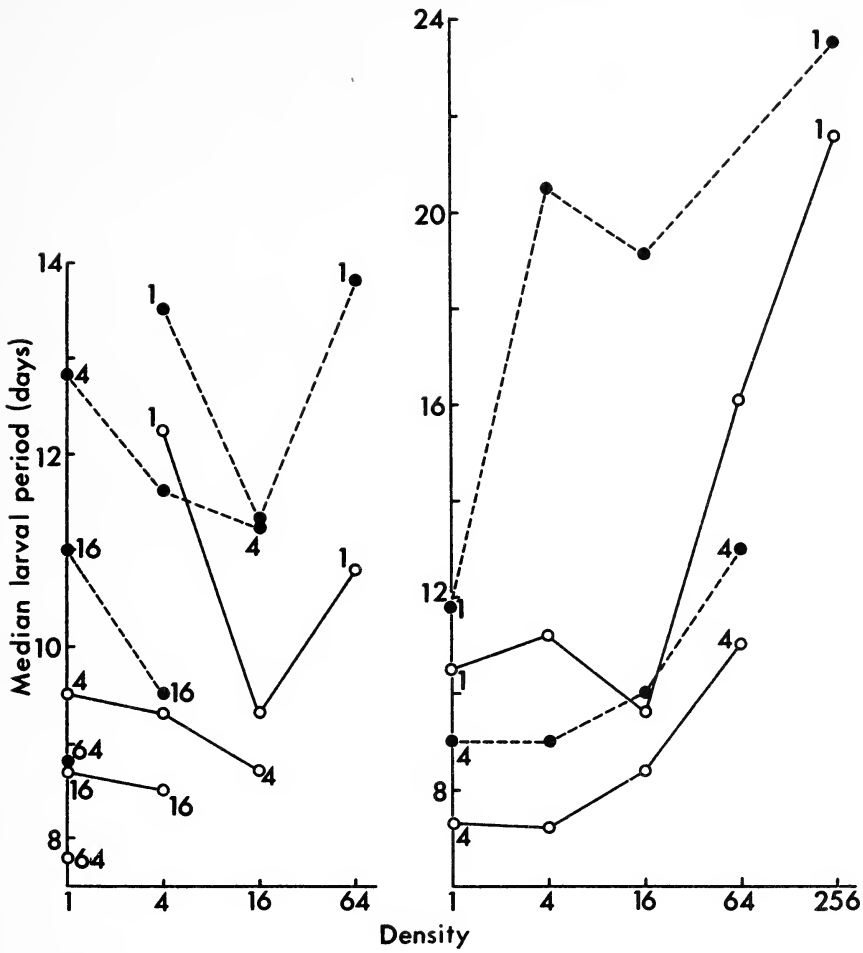
Figs. 15 & 16. Median larval period of *Aedes aegypti* at each density level.
Points for the same amount of yeast per larva are connected
by lines. 15. Experiment I. 16. Experiment IV. ○ : males;
● : females.

TABLE 7 - The ratio of larval to pupal period of the females of
         Aedes aegypti(Experiments I - IV).

Density	Food used*	Exp. I	Exp. II	Exp. III	Exp.IV	Mean
1	Y1				4.88	4.88
	Y4	4.60			3.72	4.16
	Y16	3.96				3.96
	Y64	3.17				3.17
	Y64 + R100		2.84			2.84
4	Y4	4.86			8.47	6.62
	Y16	4.17			3.72	3.95
	Y64	3.42				3.42
	Y64 + R100		2.91			2.91
16	Y4	**				**
	Y16	4.06			7.93	6.00
	Y64	4.06		3.80	4.09	3.98
	R100			2.94		2.94
	Y64 + R100		3.02			3.02
	Y64 + R200			2.88		2.88
64	Y4	**				**
	Y16	**				**
	Y64	4.96			**	>4.96
	Y256				5.37	5.37
	Y64 + R100		3.25			3.25
128	Y64 + R100		4.12			4.12
256	Y256				9.75****	9.75***
	Y1024				5.45***	5.45***

* See Table 3.
, * See Table 6.
**** Only three females pupated.

        In Fig. 21, the relation between mean wing length and mean
thorax length is shown for each density level in Experiment II and
for each type of food in Experiment III. In density level of 16 or
more in Experiment II, both wing and thorax decrease in length
along a straight line with increasing density. However, in densities
lower than 16, decreased wing length and rather unchanged thorax
length are shown, that is, the points for density levels of 1 and 4
are situated above the line through the points for density 16 to 128.
The larvae in lower density receive relatively large amounts of
food, because the amount of food per cup was kept constant in this

experiment. Therefore, it may be said that at these low density
levels the adults resulting from favorable conditions have relatively
shorter wing length than those from less favorable conditions.

The same is seen in Experiment III, where different diets
were given to the larvae with the same density of 16. The point for
the adults from the culture containing yeast 64 units plus rabbit
pellets 200 units, which is more suitable than yeast 64 plus rabbit
pellets 100 used in Experiment II, is situated above the line through
the points for density 16 to 128 in Experiment II, and the point for
the less suitable diet, yeast 64, below the line.
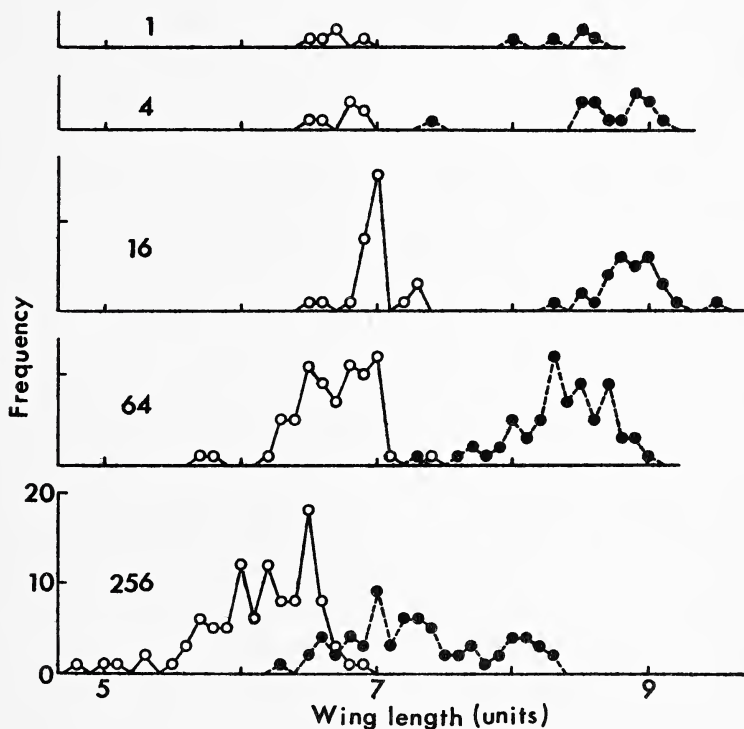
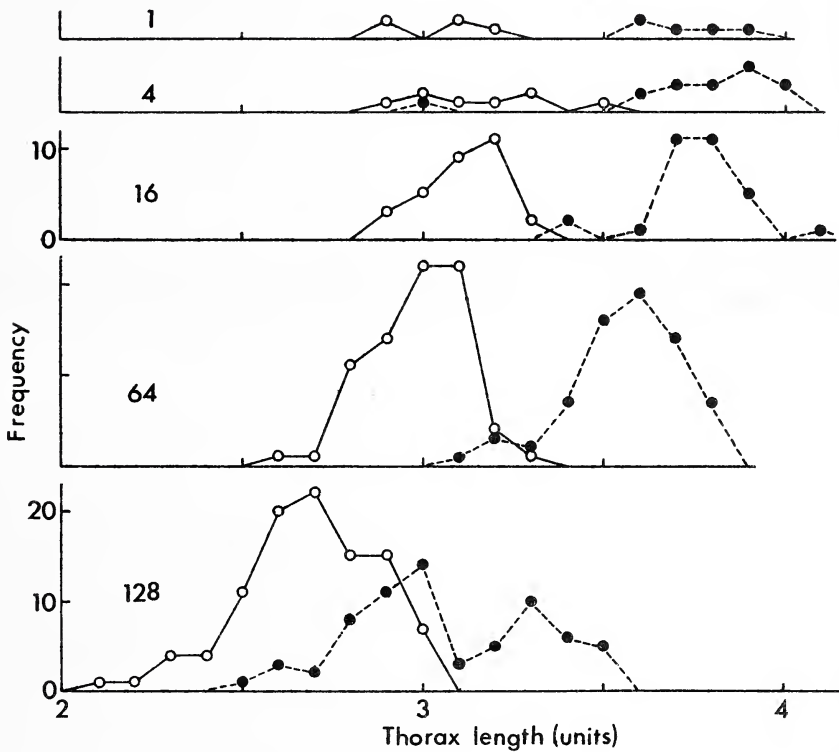Fig. 17. Frequency distributions of wing length of *Aedes aegypti* (Experi-
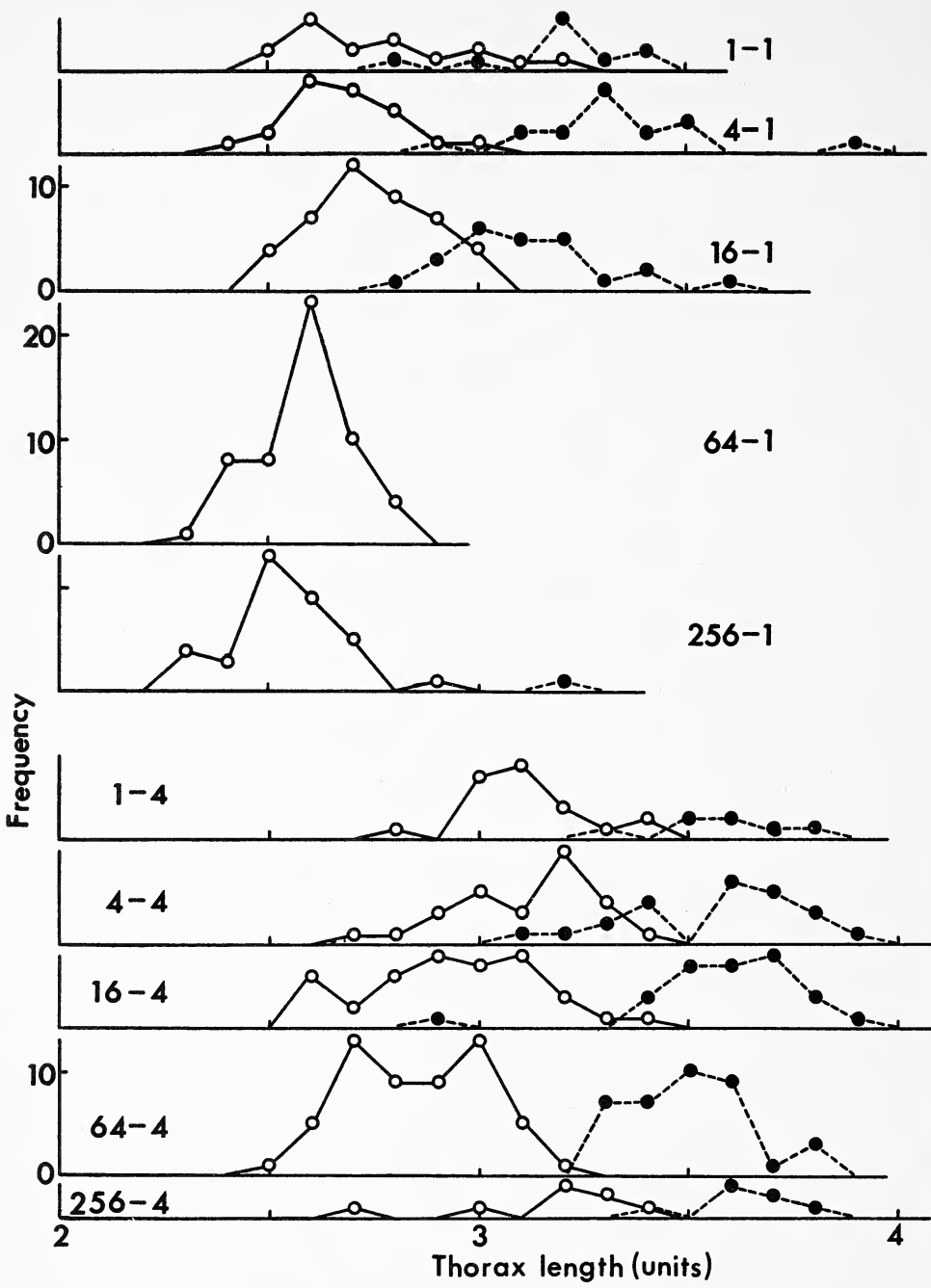ment II). Figure indicates larval density.   O : males;
● : females.
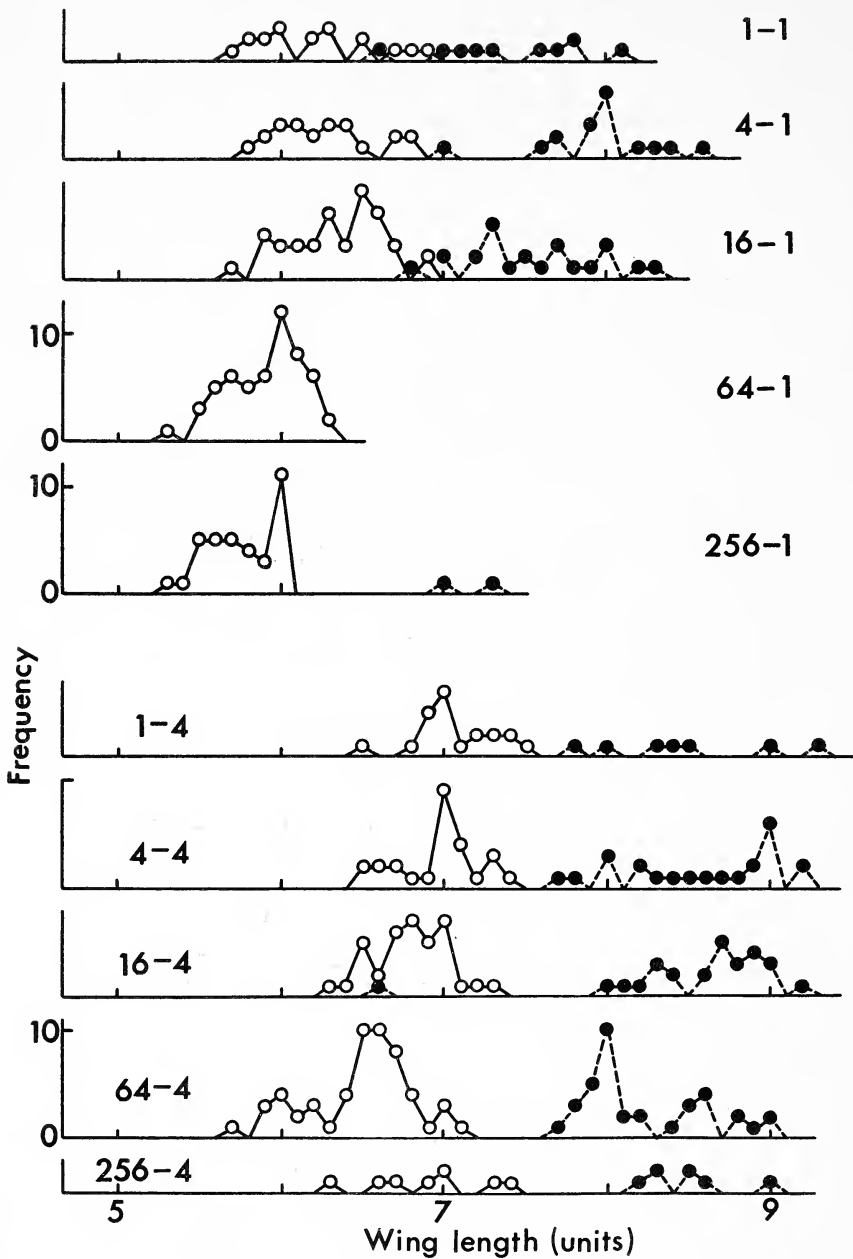
Fig. 18. Frequency distributions of wing length of *Aedes aegypti* (Experiment IV). 16-4, for example, indicates that the larval density is 16 and the amount of yeast is 4 units per larva. ○ : males; ● : females.

In Experiment IV, the situation becomes more complicated, because the experiment consisted of two series of constant amount of food per larva, and it is not easy to say which combination of larval density and amount of food is more favorable for the larval stage, especially at lower density levels. However, it is seen that the relative wing length to thorax length for larger amounts of food or lower density of larvae tends to be smaller than others.

Fig. 19. Frequency distributions of thorax length of *Aedes aegypti* (Experiment II). Figure shown indicates larval density. O : males; ● : females.
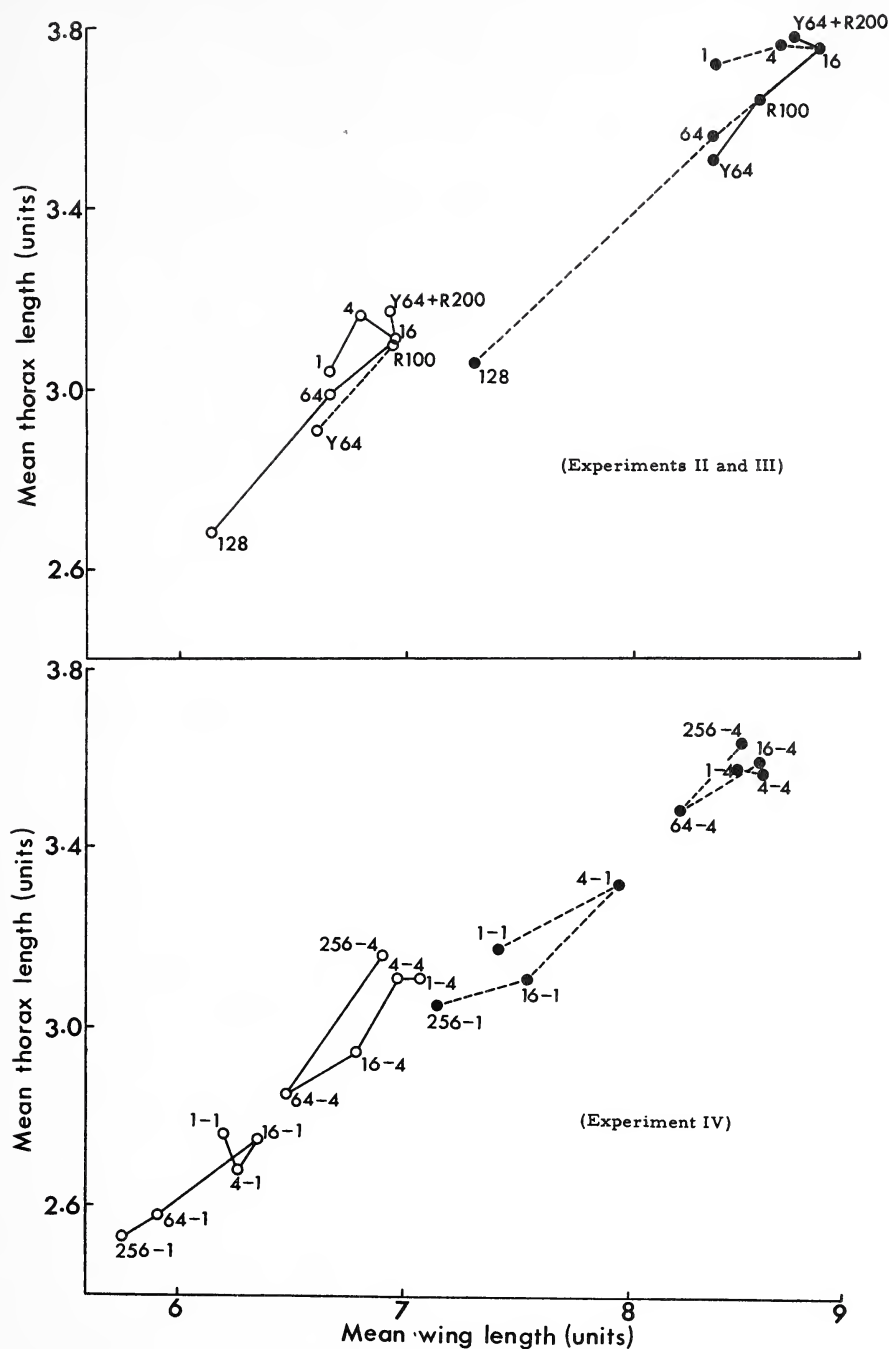
Fig. 20. Frequency distributions of thorax length of *Aedes aegypti* (Experiment IV). 16-4, for example, indicates that the larval density is 16 and the amount of yeast is 4 units per larva. ○ : males; ● : females.

Figs. 21 & 22. The relation between mean thorax length and mean wing length of
Aedes aegypti. Figure shown indicates larval density in Experiment II, and
and Y, R, and accompanied figure indicate yeast, rabbit pellets, and their
amounts in units in Experiment III. In Experiment IV, 16–4 for example,
indicates larval density – yeast units. O : males; ● : females.

The adults from the culture with density 1 and yeast 4 per cup are not considered to have a relatively smaller wing length than other densities, unlike Experiment II. This is perhaps because of the fact that the food of yeast 4 in Experiment IV is apparently less favorable than that of yeast 64 plus rabbit pellets 100. The adults with relatively small wing length from low larval density seem to appear only when the amount of food is large.

CONCLUSIONS

The results obtained are summarized in Table 8. The density in this table is used in a relative sense to food quantity. Actual density differs according to the amount of food.

TABLE 8 - Summary of the effects of larval density in *Aedes aegypti*,

Density	Larval mort.	Larval period	Variation in larval period	Sex ratio	Wing length	Thorax length	Wing/ thorax
Very low	Low	Short	Small	1/1	Large	Large	Small
Low	Very low	Very short	Small	1/1	Very Large	Large	Large
High	High	Long	Large	♂ > ♀	Small	Small	Large

High larval density apparently has detrimental effects on the mosquito. Interesting is the relation between very low and low densities. The characteristics seem to indicate that the adults from very low larval density have a slightly reduced flight ability in comparison with those from less low density, as far as judged from the relative wing length. However, repeated experiments are desired, as the number of mosquitoes used in lower densities was not very large.

CONSIDERATIONS ON THE MANNER IN WHICH
LARVAL DENSITY PRODUCES ITS EFFECTS

From the preceding sections, the effect of larval density is apparent, but its process was not particularly investigated. Since no effects of metabolic wastes of larvae have been demonstrated (Bar-Zeev, 1957; Shannon and Putnam, 1934), high larval density seems to influence the mosquitoes through the stimulation of increased mutual contacts.

Shannon and Putnam (1934) stated "DeBuck, Schoute, and Swellengrebel (1932) claim ... that when they (anopheline larvae) live in overcrowded conditions food may remain undigested in the alimentary tract from 12 to 24 hours ... Improper nourishment due to massing habits of the larvae (of *Aedes aegypti*)may account for this (phenomenon at high larval density) ...". However, the situation seems to be more complex than Shannon and Putnam (1934) thought, and neuro-physiological processes may be involved.

ACKNOWLEDGEMENTS

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CORRIGENDA

Page 101, couplet 4, add page numbers: 138 and 111.
        couplet 7, add page number 102.
Page 109, lines 15 & 16, insert:
Distribution
        A total of 3,433 specimens was examined.
Page 151, caption for Fig. 37 should read:
        C. oregona X *C. duodecimguttata* from Garth, Alberta.
        Add caption for Fig. 38:
        C. oregona X *C. duodecimguttata* from Rocky Mountain House, Alberta.
Page 169, after MacGinitie, insert:
Martin, P.S. 1958. Pleistocene ecology and biogeography of North America, pp. 375-420, *in* C.L. Hubbs, (ed.), Zoogeography. Amer. Assoc. Adv. Sci. Symp., Washington, D.C.

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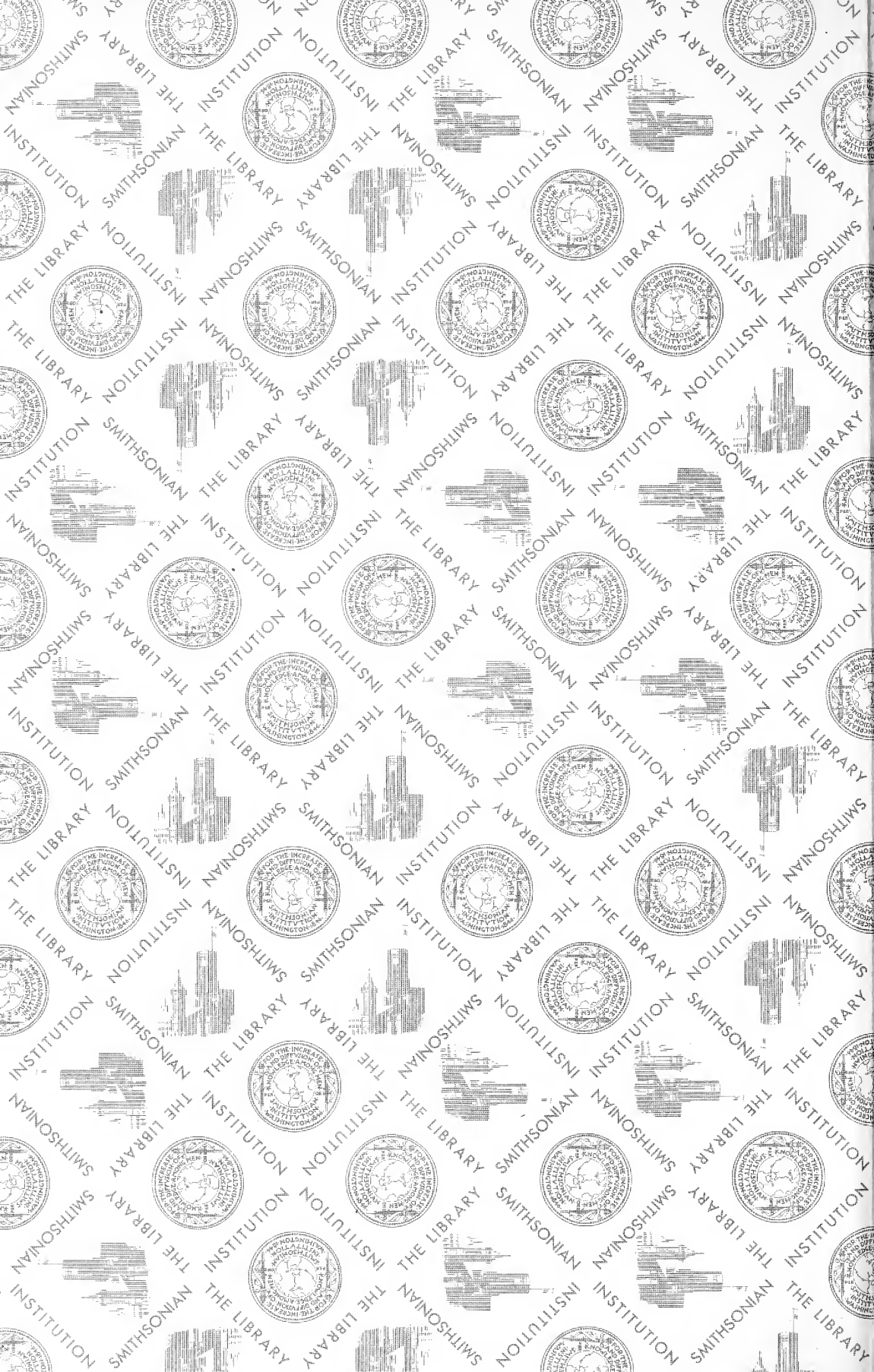
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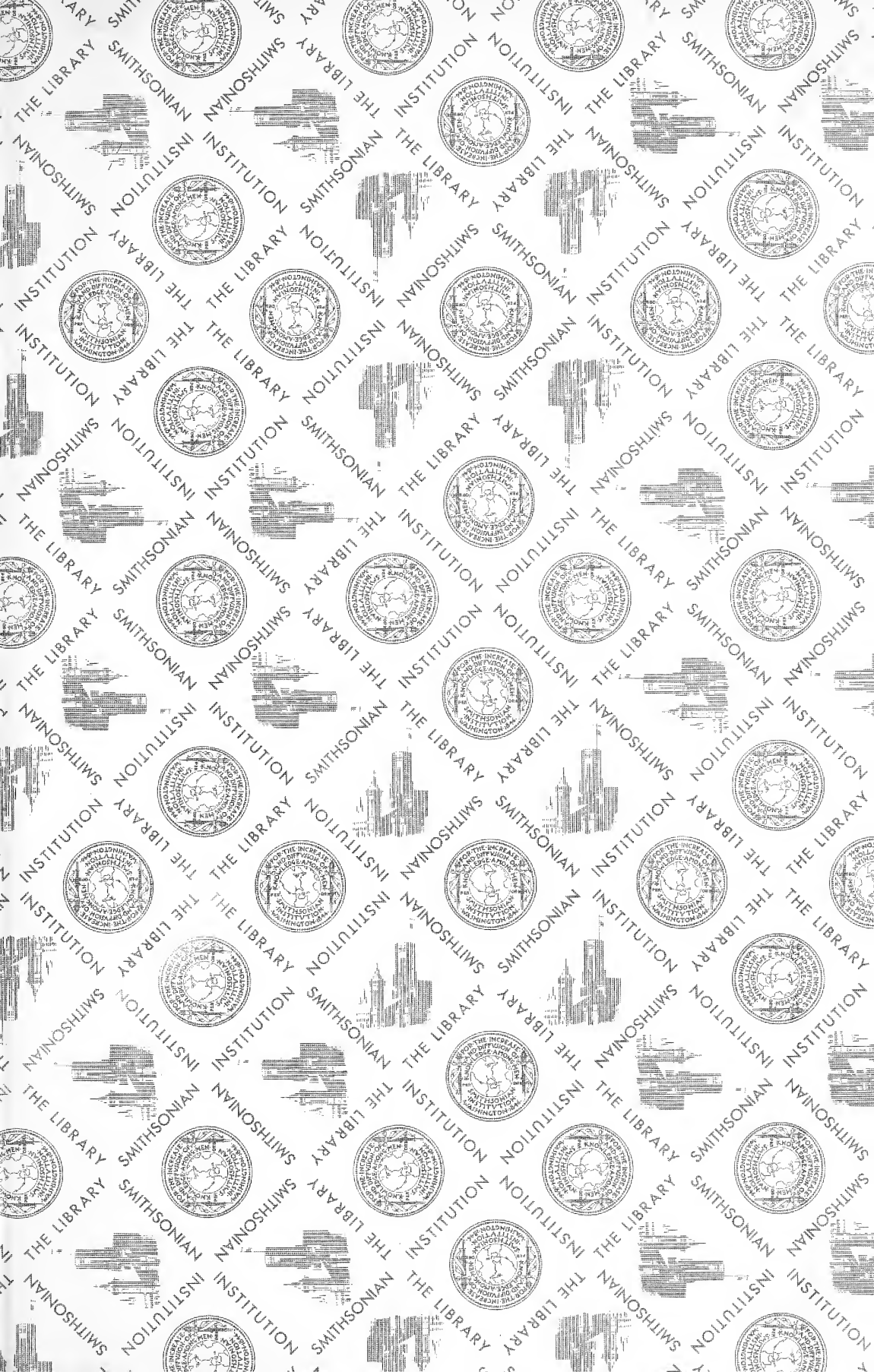
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